Reports *of*

Experiments Referred to at Hearings on Ice Cream

Before Dr. C. L. Alsberg
Chief of the Bureau of Chemistry
United States Department
of Agriculture

Subject:

The Bacteriology of Ice Cream

Published by

THE NATIONAL ASSOCIATION OF
ICE CREAM MANUFACTURERS

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FOREWORD

In June, 1914, the National Association of Ice Cream Manufacturers published the Report of the hearings held before Dr. C. L. Alsberg on the use of colloids as stabilizers in ice cream, the butter fat standard for ice cream and the bacteriology of ice cream, and in that part of the report which relates to the bacteriology of ice cream it appears that at the hearing Professor Gordon, Professor Prescott, Dr. Heinemann and Dr. Pease repeatedly referred to experiments which they had made and upon which they based their testimony. The National Association of Ice Cream Manufacturers now makes public the reports of these experiments for the benefit of those engaged in the ice cream industry and others interested in the subject.

These reports should be read in connection with the testimony above referred to.

Walter Jeffreys Carlin.

JOHN GORDON, graduate of the University of Missouri, formerly assistant Professor of Bacteriology, Iowa State College. Worked on dairy farms in Missouri, Iowa, Illinois and New York. Operated bottling plant for the Walker-Gordon Milk Co. Since 1911 in charge of the sanitation work of a number of plants in New York State, purchasing several million pounds of milk yearly. Author of a number of papers on the subect of sanitation of dairy plants, etc.

SAMUEL C. PRESCOTT, Professor of Industrial Micro-biology, Massachusetts Institute of Technology. Graduate of the Massachusetts Institute of Technology, class of 1904. Formerly Assistant Chemist and Bacteriologist of the Worcester Sewerage Works. Studied in Denmark and Germany, devoting time especially to bacteriology and fermentation in food. Since 1905 director of the Boston Bio-Chemical Laboratory. Joint author of work on bacteriology and also a book dealing with canning. Connected with the Journal of Bacteriology and author of numerous papers dealing with scientific subjects. Member of the Society of American Bacteriologists and Fellow of the American Society for the Advancement of Science and member of the American Chemical Society, the British Society of Chemical Industry, American Public Health Association and the Boston Society of Medical Sciences.

P. G. HEINEMANN, Bachelor of Science. Doctor of Philosophy, University of Chicago. Instructor in Bacteriology, University of Chicago, and Serum Expert of the Memorial Institute for Infectious Diseases, Chicago. Publisher of many pamphlets dealing with different phases of the milk question, also a book on bacteriology. Reviewer of important scientific papers for the German publications on bacteriology. Editorial writer for the Journal of the American Medical Association. For two years examiner and inspector for the Chicago Medical Milk Commission.

HERBERT D. PEASE, Doctor of Medicine, University of Toronto. Post-graduate student and fellow-in pathology, Johns Hopkins University. For one year resident bacteriologist and pathologist of the Thomas Wilson Sanitarium for Sick Children. For four years First Assistant Bacteriologist of the Bureau of Health of the City of Philadelphia. For two years bacteriologist of the New York State Pathological Laboratory. For one year instructor in bacteriology in the Sheffield Scientific School, Yale University. For nine years director of the Hygienic and Antitoxin Laboratory of the New York State Department of Health. At present and for four years past director of the Department of Bacteriology of the Lederle Laboratories of New York City. At present and for three years past sanitary expert of the Board of Water Supply of the City of New York.

Report on Ice Cream Examinations Outlined in Washington Hearing of Ice Cream Manufacturers

JOHN GORDON

REPORT ON ICE CREAM EXAMINATIONS OUTLINED IN WASHINGTON HEARING OF ICE CREAM MANUFACTURERS.

John Gordon.

The experimental work as set forth herein was carried on with a view of obtaining exact information as to the factors which influence the bacteria content of ice cream as manufactured in commercial plants. The raw materials for ice cream manufacture were followed from the place of origin, the farm, through the creamery and condensory into the ice cream factory. In the creamery and condensory the effect of the handling consequent to the ordinary factory routine involved in the receiving, separating, pasteurizing, condensing, etc., of milk upon the bacteria content of the raw materials was considered. In the ice cream factory the effect of all routine operations upon the bacteria content of the ice cream up until the time of delivery to the retailer were considered.

Methods.

The agar employed in all experiments reported herein was prepared after the method recommended by the American Public Health Associations' Committee on Standard Milk Analysis. All plates were incubated at 37° C. for a period of 48 hours.

Since most of the ice cream experiments were made on semi-hard ice cream, samples were taken directly from the freezers in sterile bottles holding seventy cubic centimeters, which were stoppered and carried immediately to the laboratory of the Wheat's Ice Cream Company, wherein most of the work was done.

When it was necessary to sample hard ice cream in the containers, this was done by forcing a sterile butter trier down through the ice cream in the can. After the core thus obtained was withdrawn, it was broken with a sterile spatula and except for a thin section on the top was

placed in a sterile wide-mouthed sample bottle, holding seventy cubic centimeters.

After the samples had reached the laboratory, they were in all cases melted by placing the bottles in a dish containing warm water. After melting, the routine procedure of an ordinary milk analysis was followed.

Many of the tables given are a fractional part of the data on that particular subject, being the results of the routine tests of a large commercial ice cream manufactory. Naturally, nothing would be gained by printing the results of a great many routine tests where a few will depict average conditions satisfactorily.

Tests on Farm Milks Received At a Creamery Supplying Several Ice Cream Factories.

THE BACTERIAL CONTENTS OF FARMERS' MILKS AT TIME OF DELIVERY TO CREAMERY ON SUCCESSIVE DAYS.

Sample	Patron	Bacteria Content Per C.C.
1	No. 170	1,700,000
2	" 61	6,100,000
3	" 154	15,400,000
4	" 285	90,000
5	" 285	55,000
6	" 77	32,500
7	" 77	2,500,000
8	" 265	2,850,000
9	" 265	280,000
10	" 15	950,000
11	" 15	155,000
12	" 15	1,250,000
13	" 266	7,000,000
14	" 266	28,000
15	" 215	2,000,000
16	" 281	960,000
17	" 281	3,000,000
18	" 224	700,000
19	" 224	110,000
20	" 224	450,000
21	" 292	480,000
22	" 292	10,000,000
23	" 292	1,800,000
24	" 66	820,000
25	" 101	95,000
26	" 101	120,000
27	" 288	360,000
28	" 288	1,750,000
29	" 288	200,000

The above table is used merely to show the general run of bacteria contents of farmers' milk as received at an ice cream creamery. It represents only a few of the tests made, but the printing of more would be needless, as it shows that one farmer may bring milk of high bacteria

content one day and milk of low bacteria content the next.

The Bacteria Content of Milk, Cream, and Condensed Milk, Used For Ice Cream Manufacture As Received At Several Ice Cream Plants.

The following two tables depict the condition in which cream arrives at the average ice cream factory from the average country creamery:

BACTERIA CONTENTS OF PASTEURIZED CREAM FROM CREAMERY A AT TIME OF DELIVERY TO ICE CREAM PLANT IN ROCHESTER, NEW YORK.

Date		Bacteria Content Per C.C.
Aug. 21st	Can 1	26,100,000
	" 2	9,500,000
	" 3	51,600,000
	" 4	52,500,000
	" 5	27,400,000
Aug. 23rd	" 1	11,500,000
	" 2	11,000,000
Aug. 24th	" 1	22,500,000
	" 2	19,600,000
Aug. 29th	" 1	29,000,000
	" 2	31,100,000
	" 3	20,000,000
	" 4	22,800,000
Sept. 1st	" 1	1,300,000
	" 2	1,450,000

BACTERIA CONTENTS OF PASTEURIZED CREAM FROM CREAMERY B SHIPPING TO THE ROCHESTER PLANT.

Date		Bacteria Content Per C.C.
Aug. 9th	Can 1	5,000,000
	" 2	10,000,000
	" 3	4,300,000
Aug. 24th	" 1	5,100,000
	" 2	15,200,000
	" 3	15,000,000
	" 4	1,500,000
Sept. 2nd	" 1	1,060,000
	" 2	1,760,000

Since I was convinced that the high counts obtained on the pasteurized cream from the creameries A and B were due not to faulty methods of pasteurization but to poor storage facilities since they, like the great bulk of creameries, relied upon the ice tank method of storage, I had one of the creameries ship cream to the ice cream factory directly after pasteurization and cooling and not hold it in the ice tank until the next day as was the usual custom. The ordinary method of pasteurization was followed, no unusual precautions being taken.

BACTERIA CONTENTS OF PASTEURIZED CREAM PROPERLY COOLED AFTER PASTEURIZATION AND SHIPPED IMMEDIATELY.

Date		Bacteria Per C.C.
Sept. 2nd	Can 1	39,000
	" 2	21,000
	" 3	71,000
	" 4	39,000
	" 5	33,000
	" 6	70,000
	" 7	35,000
	" 8	64,000
	" 9	26,000

The following table shows that the ice cream manufacturer may when he purchases pasteurized milk be obtaining milk of much higher bacteria content than raw milk. The milk in this case was sweet and apparently good but it required a bacteriological analysis before the enormous bacteria content was revealed:

BACTERIA CONTENTS OF PASTEURIZED MILK SOLD TO AN ICE CREAM COMPANY BY A DAIRY CONCERN.

Can 1	60,000,000	Bacteria per C.C.
" 2	420,000,000	" " "
" 3	21,000,000	" " "
" 4	100,000,000	" " "

THE BACTERIA CONTENTS OF HOMOGENISED CREAM FROM CREAMERY C AS RECEIVED AT AN ICE CREAM FACTORY ON DIFFERENT DAYS.

Date	Sample	Bacteria Per C.C.
Aug. 3	1	4,800
" 4	2	6,500
" 5	3	8,000
" 6	4	15,500
" 7	5	5,000
" 8	6	3,400
" 9	7	80,000
" 10	8	250,000
" 11	9	125,000
" 12	10	30,000

Since the process of homogenization as followed at this creamery required a temperature of 180° F., it followed that the bacteria content would be low. Only a very small percentage of ice cream manufacturers have homogenizers, however, therefore homogenizers cannot be a factor in reducing the bacteria content of ice cream.

BACTERIA CONTENT OF CONDENSED MILK FROM TWO CREAMERY AND CONDENSERIES.

Date	Condensery	Bacteria Per C.C.
July	A	2,940,000
"	A	50,000
"	A	53,000
Aug.	A	10,000
"	A	35,000
"	A	80,000
"	B	5,000,000
"	B	180,000
"	B	2,000,000

The foregoing shows that the bacteria content of the type of condensed (unsweetened whole milk) used in ice cream manufacture fluctuates after the fashion of normal milk or cream. The condensing temperature (120° F.) kills only a portion of the bacteria, nor is the superheating temperature (180° F.) sufficient to dispose of the entire number. Numerous factors can increase the bacteria content of condensed milk, temperature being the chief one. If the condensed milk is not cooled to a low enough temperature after being drawn from the condensing pan, the bacteria content will increase to enormous proportions. Unsweetened condensed milk must be cared for with the ordinary care awarded normal milk. The best possible storage conditions must obtain for its successful storage.

The Increase In Bacteria Contents of Raw Materials During Shipment By Rail.

It is not customary for railroads to provide ice for the preservation of milk in transit. In the hot summer weather enormous increases of the bacteria content of milk and much actual souring of milk results from the faulty methods of transportation in use.

The following table does not depict average conditions, because the creamery mentioned in the experiment is equipped with artificial refrigeration and consequently secures much lower temperatures prior to shipment than the average creamery can secure:

TABLE SHOWING INCREASE OF TEMPERATURES AND BACTERIA IN FIVE-HOUR SHIPMENT PERIOD OF MILK FROM AKRON, N. Y., TO ROCHESTER, N. Y., ON A COOL DAY IN AUGUST, MAXIMUM TEMPERATURE 76° F.

	Temperature Akron, N. Y.	Temperature Rochester, N. Y.	Increase in Temperature
Can 1	43° F.	55° F.	12° F.
" 2	44° "	55° "	11° "
" 3	44° "	52° "	8° "
" 4	42° "	53° "	11° "
" 5	44° "	55° "	11° "
" 6	43° "	53° "	10° "
" 7	45° "	55° "	10° "
" 8	46° "	54° "	9° "
" 9	45° "	54° "	9° "
" 10	45° "	55° "	10° "

BACTERIA CONTENTS OF FOREGOING CANS OF MILK AT STARTING AND FINISHING POINTS OF SHIPMENT.

	At Akron, N. Y. Bacteria Per C.C.	At Rochester, N. Y. Bacteria Per C.C.	Increase
Can 1	140,000	230,000	90,000
" 2	117,000	250,000	133,000
" 3	96,000	300,000	204,000
" 4	107,000	210,000	103,000
" 5	97,000	225,000	128,000
" 6	85,000	330,000	265,000
" 7	170,000	265,000	95,000
" 8	225,000	320,000	95,000
" 9	130,000	420,000	290,000
" 10	120,000	850,000	730,000
	Average Bact. Content 127,000	Aver. Bact. Content 342,000	Aver. Increase 210,000

The above demonstrates that even on a cool day milk shipped at a low initial temperature will very materially increase, in fact almost double in bacteria content.

Storage of Ice Cream Materials.

A SIX DAY TEST ON THE BACTERIAL GROWTH IN UNPASTEURIZED CREAM STORED AT 32°F. IN THE COLD ROOM OF AN ICE CREAM PLANT.

		Bacteria Per C.C.
First day	Can 1	12,000,000
	" 2	10,500,000
	" 3	12,500,000
Third day	" 1	12,800,000
	" 2	10,800,000
	" 3	13,000,000
Fourth day	" 1	12,000,000
	" 2	9,000,000
	" 3	10,000,000
Fifth day	" 1	14,800,000
	" 2	10,000,000
	" 3	14,500,000
Sixth day	" 1	17,800,000
	" 2	12,200,000
	" 3	13,500,000

The above table shows a material increase in bacteria in unpasteurized cream in storage at 32° F. starting on the sixth day. This is a longer period than cream is commonly stored in the ice cream factory.

It should be borne in mind that only a small percentage of ice cream manufacturers have artificial refrigeration, therefore, the best temperature of storage obtainable by many ice cream factories is 40° F. to 50° F. and their

milk and cream storage facilities are confined to ice tanks or ice boxes.

The Bacteria Contents of Other Ice Cream Materials.

The following gelatine analyses were made by weighing out one gram of gelatine on a sterile filter paper, which was afterwards used funnel fashion to pour the gelatine into a 99 cubic centimeter water blank which had been warmed up slightly. After the warm water had taken the gelatine into solution the bottle was considered to contain a 1-100 dilution and the test proceeded with accordingly:

BACTERIA CONTENTS OF RAW GELATINE AS RECEIVED FROM MANUFACTURER OR JOBBER.

Sample	Bacteria Content Per Gram	Sample	Bacteria Content Per Gram
1	1,150,000	14	29,000,000
2	2,050,000	15	1,600,000
3	2,850,000	16	20,000,000
4	45,000	17	28,500,000
5	2,370,000	18	25,000,000
6	200	19	11,000,000
7	3,500	20	16,000,000
8	4,000	21	600
9	20,000,000	22	1,200
10	30,000,000	23	14,000
11	75,000	24	200
12	30,000	25	200
13	200,000	26	100

The last six samples are table gelatines. The preceding samples are ice cream gelatines.

The Preparation of Raw Gelatine For Use In Ice Cream.

This is properly done by bringing the desired amount of water to the point where it boils vigorously and then shutting the steam off. The gelatine should then be poured into the cooker. A large part of it will immediately dissolve and a slight stirring will cause the rest to do the same. The gelatine is immediately poured into the ice cream. This process, which is followed in the modern plants, secures very satisfactory results and renders certain the almost complete destruction of the bacteria in the raw gelatine. The bacteria content of gelatine prepared in this fashion is rarely over 2,000 bacteria per cubic centimeter.

Bacteria Content of Sugar.

Analyses of sugar secured from several ice cream

plants were practically negative in that they contained practically no bacteria. Molds, however, were occasionally present.

BACTERIA CONTENT OF VANILLA.

Sample	Bacteria Per C.C.	Sample	Bacteria Per C.C.
1	1,800	3	100
2	1,700	4	2,000

Effect of Mixing And Freezing Processes on the Bacteria Content of Ice Cream.

It is a generally recognized fact that agitation of a milk sample produces an apparent increase of bacteria in the same, due to the breaking up of clumps, which, had they remained intact, would have produced only one colony when plated. In cream there is an added incentive toward the clumping of bacteria and a consequent uneven distribution, owing to its heavy, viscid nature.

Experiments made at the Iowa Experiment Station while the writer was working on the effect of freezing on Lactic Acid bacteria in "Lacto," a frozen product made of clabbered milk, and originated at that Station, revealed an enormous apparent increase in these bacteria in the frozen lacto as compared with the unfrozen material. Much of the increase I ascribed to the breaking up of clumps of coagulated milk, although prior to freezing, the mixed material was carefully strained through a very fine meshed strainer. How much of the increase was due to the breaking up of aggregates of bacteria, I would not attempt to say.

After noticing on many occasions a marked increase in the bacterial content of the frozen ice cream as made in commercial plants over the mixed materials, I concluded that the increase was due to the agitation received in the mixer or in the freezers.

INFLUENCE OF MIXING PROCESS ON BACTERIAL CONTENT OF ONE HUNDRED AND SEVENTY-FIVE GALLONS OF ICE CREAM MIXTURE.

Experiment	Average Number of Bacteria before Mixing	Average Number of Bacteria after Freezing
1	22,500,000	14,500,000
2	19,000,000	20,000,000
3	25,500,000	26,500,000
4	30,000,000	28,000,000
5	39,000,000	38,000,000

From these tests it can be concluded that the agitation of the paddles in the mixer is not sufficient to create an apparent bacterial increase in the materials being mixed. This condition, however, can be brought about by holding the materials in the mixer for a much longer period of time than is usually the custom.

Effect of Agitation In Freezer on Bacteria Content of Ice Cream.

All the experiments following were made under practical conditions except that the hopper from which the unfrozen sample was taken and the freezer through which the cream passed were made practically sterile by the use of boiling hot water. The time of freezing (six to eight minutes) and the freezing temperature (10° F.) were not changed from the daily routine. All results are the averages of the results on two or more samples.

	Bacteria Per C.C. in Unfrozen Mix.	Bacteria Per C.C. in Ice Cream	Increase of Bact. Per C.C.	Decrease of Bact. Per C.C.
Test 1	13,725,000	22,900,000	9,175,000	
2	16,450,000	25,400,000	8,950,000	
3	2,925,000	2,255,000		670,000
4	2,125,000	1,950,000		175,000
5	7,000,000	7,330,000	330,000	
6	3,000,000	7,230,000	4,230,000	
7	3,500,000	5,500,000	2,000,000	
8	860,000	1,160,000	300,000	
9	725,000	1,630,000	900,000	
10	720,000	2,310,000	1,590,000	
11	5,000,000	6,600,000	1,600,000	
12	5,400,000	5,870,000	470,000	
13	1,800,000	1,413,000		400,000
14	1,675,000	1,750,000	75,000	
15	1,950,000	2,120,000	170,000	
16	4,900,000	7,500,000	2,600,000	
17	3,650,000	6,200,000	2,550,000	
18	7,500,000	15,000,000	7,500,000	
19	5,700,000	10,500,000	5,000,000	
20	1,200,000	1,700,000	500,000	
21	6,500,000	15,000,000	8,500,000	
22	8,500,000	12,000,000	3,500,000	
23	13,200,000	13,000,000		200,000
24	9,500,000	17,800,000	8,300,000	
25	11,200,000	12,500,000	1,300,000	
26	530,000	650,000	130,000	
27	200,000	240,000	40,000	
28	600,000	700,000	100,000	
29	370,000	570,000	270,000	
30	150,000	650,000	500,000	
31	170,000	520,000	350,000	

The above results readily prove that the rapidly revolving dashers in the ice cream freezer, by their action upon clumps of bacteria produce an apparent increase in the total number of bacteria in the frozen ice cream.

Increase Experiment In Which All the Materials of a Batch of Ice Cream Were Tested Prior to Mixing.

MILK.

	Bacteria Per C.C.		Bacteria Per C.C.
Can 1	10,000,000	Can 4	10,000,000
" 2	6,000,000	" 5	140,000,000
" 3	9,000,000		

CREAM.

	Bacteria Per C.C.		Bacteria Per C.C.
Can 1	40,000,000	Can 4	6,000,000
" 2	2,000,000	" 5	1,500,000
" 3	2,700,000	" 6	

CONDENSED MILK.

	Bacteria Per C.C.		Bacteria Per C.C.
Can 1	40,000,000	Can 3	10,000,000
" 2	7,000,000	" 4	

OTHER MATERIALS.

Gelatine—1,000 bacteria per cubic centimeter.
Sugar—No bacteria; a few molds.
Vanilla—100 bacteria per cubic centimeter.

BACTERIAL CONTENT OF MIXED MATERIALS IN MIXER.

Average of two samples—25,500,000 bacteria per cubic centimeter.

BACTERIAL CONTENT OF MIXED MATERIALS TAKEN FROM HOPPER ON TOP FREEZER.

Freezer No. 14	Freezer No. 15
26,500,000	26,000,000

BACTERIAL CONTENT OF THE FROZEN ICE CREAM.

Freezer No. 14	Freezer No. 15
40,500,000	46,500,000

EFFECT OF STORAGE TEMPERATURES ON BACTERIAL CONTENT OF ICE CREAM MADE IN ABOVE EXPERIMENT IN FREEZERS 14 AND 15 FROM TESTED MATERIALS. AVERAGE STORAGE TEMPERATURE, ZERO FAHRENHEIT.

	Freezer 14		Freezer 15
Storage	Bacteria Per C.C.	Storage	Bacteria Per C.C.
1st day	40,500,000	1st day	46,500,000
3rd "	42,000,000	3rd "	39,500,000
4th "	18,500,000	4th "	19,000,000
5th "	21,500,000	5th "	26,000,000
6th "	35,000,000	6th "	28,000,000
8th "	36,000,000		
10th "	37,000,000	10th "	33,000,000
14th "	21,800,000	14th "	14,700,000
20th "	16,500,000		

Disregarding the rise and fall of the number of bacteria from day to day due to unequal distribution, it can be seen that there is a gradual decline of the number of bacteria in the stored ice cream beginning with the first day of storage.

EXPERIMENT SHOWING EFFECT OF ZERO FAHRENHEIT STORAGE TEMPERATURES ON ICE CREAM OF LOW BACTERIA CONTENT.

	1st Day	8th Day
Can 1	55,000	52,000
" 2	49,000	42,000
" 3	55,000	48,000

A slight steady decline in the numbers of bacteria is apparent in the above results.

Distribution of Bacteria In Ice Cream.

BACTERIAL CONTENTS OF VARIOUS SAMPLES FROM THE CONTENTS OF TEN-GALLON ICE CREAM FREEZERS.

Sample	Freezer 1 Bacteria Per C.C.	Sample	Freezer 2 Bacteria Per C.C.
1	20,000,000	1	21,400,000
2	24,000,000	2	25,500,000
3	14,000,000	3	29,600,000
4	23,000,000	4	27,000,000
5	16,500,000	5	25,000,000
6	20,000,000	6	24,000,000
7	31,000,000		
8	23,700,000		
9	29,500,000		
10	27,600,000		

BACTERIA CONTENTS OF SAMPLES FROM VARIOUS PORTIONS OF A CAN OF ICE CREAM.

Sample	Can 1 Bacteria Per C.C.	Sample	Can 2 Bacteria Per C.C.
1	7,600,000	1	14,500,000
2	9,500,000	2	21,000,000
3	13,500,000	3	16,000,000
4	17,500,000	4	18,500,000
5	9,000,000	5	14,000,000

Can 1	Can 2
Widest Variation 9,900,000 Bacteria Per C.C.	Widest Variation 7,000,000 Bacteria Per C.C.

The results in this and the preceding table show that the distribution of bacteria in ice cream is markedly uneven.

The following bacteriological analyses of the product of a large commercial ice cream plant operated in a most cleanly fashion, show that such plants turn out ice cream of high bacteria content despite their many precautions. It can also be seen that the bacteria content of the ice cream varies greatly, both on the same day and succeeding days:

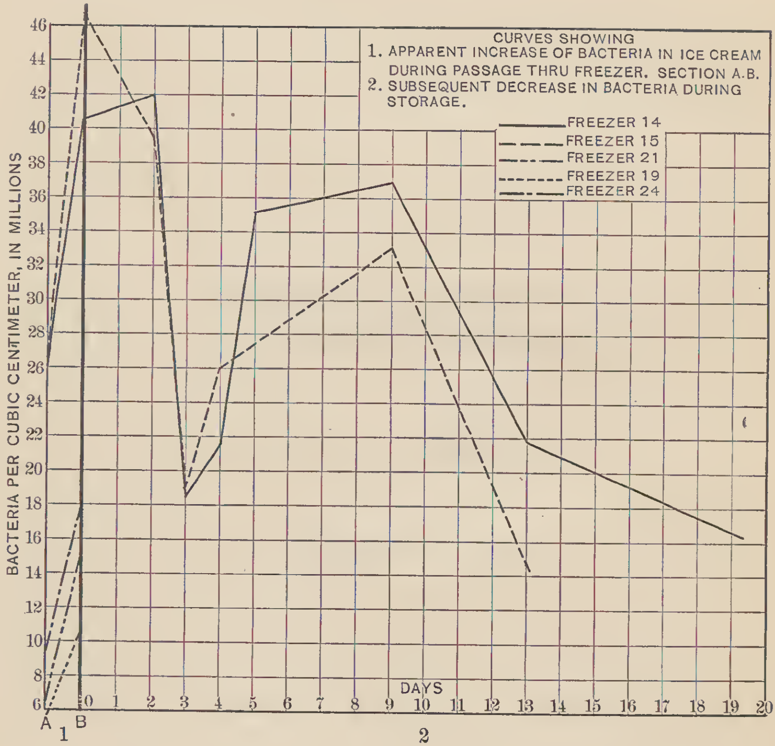
It can safely be said that of the numbers of bacteria recorded in the following table that only an infinitesimal percentage were due to the utensils in the ice cream plant.

The ice cream was positively manufactured under the best conditions obtainable in the ice cream industry to-day:

BACTERIA CONTENTS OF ICE CREAM MADE ON SUCCESSIVE DAYS IN A SANITARY ICE CREAM PLANT.

Date	Sample	Bacteria Per C.C.
August 11th	1	11,000,000
	2	10,000,000
	3	14,000,000
	4	16,000,000
	5	15,000,000
	6	20,000,000
" 12th	1	5,000,000
	2	20,000,000
	3	15,000,000
	4	19,000,000
" 13th	1	1,600,000
	2	2,500,000
	3	1,500,000
	4	3,000,000
" 14th	1	10,000,000
	2	12,000,000
	3	11,000,000
	4	16,000,000
	5	2,500,000
	6	1,500,000
	7	3,000,000
" 15th	1	1,200,000
	4	1,000,000
	6	3,000,000
" 16th	1	
	2	10,000,000
	4	8,000,000
	5	5,000,000
	7	11,000,000
" 18th	1	20,000,000
	2	17,000,000
	4	16,000,000
	6	13,500,000
	7	14,000,000
	8	15,500,000
" 19th	2	10,000,000
	3	20,000,000
	4	18,000,000
	7	14,000,000
" 20th	1	12,500,000
	2	25,000,000
	4	18,000,000
	5	20,000,000
	6	22,000,000
	7	17,000,000
	8	15,000,000
" 22nd	3	4,000,000
	4	5,000,000
	5	5,500,000
	6	6,000,000
	7	4,000,000

Given raw materials of low bacteria content it can be said in absolute assurance that the ice cream manufacturer can produce with great ease ice cream of low bacteria content.


CURVES SHOWING
1. APPARENT INCREASE OF BACTERIA IN ICE CREAM DURING PASSAGE THRU FREEZER. SECTION A.B.
2. SUBSEQUENT DECREASE IN BACTERIA DURING STORAGE.
FREEZER 14
FREEZER 15
FREEZER 21
FREEZER 19
FREEZER 24
BACTERIA PER CUBIC CENTIMETER, IN MILLIONS
DAYS
6
8
10
12
14
16
18
20
22
24
26
28
30
32
34
36
38
40
42
44
46
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20
A
B
1
2

Report on Ice Cream Examinations

S. C. PRESCOTT

PROF. S. C. PRESCOTT'S REPORT.

Boston, March 4, 1914.

Walter J. Carlin, Esq.,
2 Rector St., New York City.

Dear Sir.—Attached hereto are the figures for the examination of two lots of ice cream—one a very high grade product, obtained from one of our best caterers, the other a commercial sample manufactured by one of the largest ice cream dealers in the city. These have been examined with reference to the number of bacteria developing at room temperature and body temperature upon several different media as indicated, and we have used in this investigation two different lots of media, made in exactly the same way and differing only in that one of them has been kept in storage in our ice-chest for a period of a week longer than the other. The age at the time of use was for the media marked "old" ten days, and for the media marked "new," three days. The examinations have been carried out in three different dilutions in order to observe what variations, if any, may be traced merely to difference in dilution of the sample.

The plots following the tables are in each case, I believe, self-explanatory, and have been made using the figures in each case which judgment would dictate to be the most reliable—namely—those dilutions which give from 25 to 200 or 300 bacteria per plate. The samples marked A_1, A_2, A_3 were all taken from the same general

location, and might be regarded as each forming a fraction of the top portion of the freezer. The sample marked B was taken from a point lower down in the freezer; C from a still lower layer, and D from the bottom. In this way we can determine differences, if any exist, in the portion of the freezer used. I think you will note both from the tables and the plots that it would be practically impossible to select any particular spoonful which could be taken as representing the whole mass, and you will also find that the age of the medium sometimes reacts one way and sometimes the other, but that in general the age is largely in favor of the newer or fresher medium.

Very truly yours,

Samuel C. Prescott.

EXAMINATION OF VANILLA ICE CREAM. LOT 1, CATERER.

Determination of Effect of Different Media, Different Dilutions and Different Temperatures of Incubation on Numbers of Bacteria.

SAMPLE A1.

Bacteria per c.c.

Medium	Temp.	Dilution 1 :10,000	Acid	Dilution 1 :100,000	Acid	Dilution 1 :1,000,000	Acid
New Meat agar	37°	51		4		0	
		78		spr.		2	
	20°	192		21		6	
		121		19		1	
Old Meat agar	37°	52		2		0	
		61		4		3	
	20°	184		29		spr.	
		240		19		3	
New Extract agar	37°	46		3		0	
		53		2		2	
	20°	136		24		2	
		164		25		3	
Old Extract agar	37°	32		1		0	
		38		0		3	
	20°	138		16		3	
		128		23		2	
New Casein agar	37°	19		2		0	
		23		2		16	
Old Casein agar	37°	22		6		17	
		19		5		5	
New Dextrose	37°	40	40	1	1	spr.	..
Litmus agar		spr.		..		0	0
Old Dextrose	37°	11	11	4	4	spr.	..
Litmus agar		3	3	1	0	spr.	..
New Lactose	37°	12	11	0	0	0	..
Litmus agar		2	2	spr.	0	spr.	..
Old Lactose	37°	spr.	..	0	0	0	0
Litmus agar		spr.	..	0	0	1	0
New Bile	37°	5 neg.		5 neg.		5 neg.	
Old Bile	37°	5 neg.		5 neg.		5 neg.	

SAMPLE A2.

Bacteria per c.c.

Medium	Temp.	Dilution 1 :10,000	Acid	Dilution 1 :100,000	Acid	Dilution 1 :1,000,000	Acid
New Meat agar	37°	spr.		6		spr.	
		72		10		0	
	20°	360		spr.		4	
		199		20		8	
Old Meat agar	37°	spr.		9		2	
		71		108		2	
	20°	224		32		3	
		145		26		3	
New Extract agar	37°	63		6		0	
		40		7		1	
	20°	184		29		6	
		237		spr.		7	
Old Extract agar	37°	52		3		1	
		156		7		2	
	20°	171		18		6	
		169		22		2	
New Casein agar	37°	29		10		5	
		25		0		1	
Old Casein agar	37°	62		2		0	
		30		2		1	
New Dextrose	37°	25	25	spr.	..	0	..
Litmus agar		32	32	9	9	1	1
Old Dextrose	37°	55	55	4	4	0	0
Litmus agar		spr.	..	3	3	1	1
New Litmus	37°	24	20	5	5	0	0
Lactose agar		19	19	3	3	0	0
Old Litmus	37°	22	22	3	1	2	0
Lactose agar		35	23	8	7	1	1
New Bile	37°	5 neg.		5 neg.		5 neg.	
Old Bile	37°	5 neg.		5 neg.		5 neg.	

SAMPLE A3.

Medium	Temp.	Bacteria per c.c. Dilution 1:10,000	Acid	Dilution 1:100,000	Acid	Dilution 1:1,000,000	Acid
New Meat agar	37°	63		8		spr.	
		spr.		8		4	
	20°	151		spr.		5	
		284		40		spr.	
Old Meat agar	37°	spr.		27		4	
		39		16		8	
	20°	315		30		6	
		spr.		34		5	
New Extract agar	37°	27		3		0	
		46		0		0	
	20°	132		27		.3	
		165		28		6	
Old Extract agar	37°	30		4		3	
		12		8		0	
	20°	156		27		7	
		130		spr.		spr.	
New Casein agar	37°	74		11		2	
		56		2		0	
Old Casein agar	37°	43		9		44	
		55		5		8	
New Litmus	37°	spr.	..	2	2	2	1
Dextrose agar		36	35	3	3	0	0
Old Litmus	37°	spr.	..	spr.	..	spr.	..
Dextrose agar		73	73	5	5	spr.	..
New Litmus	37°	79	78	9	7	0	0
Lactose agar		69	68	3	1	0	1
Old Litmus	37°	63	60	4	3	0	0
Lactose agar		49	49	14	14	2	2
New Bile	37°	4 neg.		4 neg.		4 neg.	
		1 pos. 15 p. c.		1 pos. 5 p. c.		1 pos. 5 p. c.	
Old Bile	37°	5 neg.		5 neg.		5 neg.	

SAMPLE B.

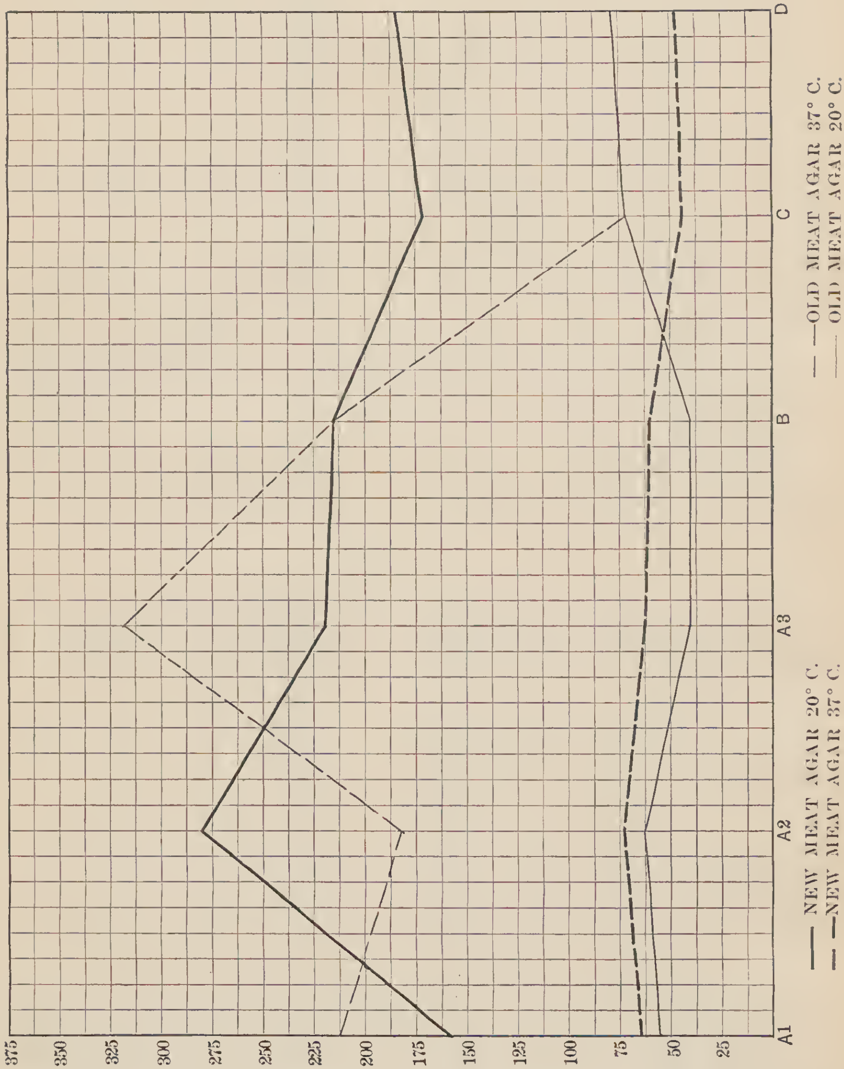
Medium	Temp.	Bacteria per c.c. Dilution 1:10,000	Acid	Dilution 1:100,000	Acid	Dilution 1:1,000,000	Acid
New Meat agar	37°	61		8		5	
		60		7		4	
	20°	215		25		spr.	
		spr.		30		spr.	
Old Meat agar	37°	spr.		spr.		1	
		39		spr.		1	
	20°	spr.		spr.		7	
		216		spr.		5	
New Extract agar	37°	5		10		11	
		8		6		10	
	20°	140		23		1	
		156		16		6	
Old Extract agar	37°	35		5		6	
		19		44		3	
	20°	141		24		7	
		135		26		7	
New Casein agar	37°	60		10		5	
		80		16		11	
Old Casein agar	37°	37		7		1	
		34		4		2	
New Litmus	37°	47	47	7	7	2	0
Dextrose agar		56	56	4	4	1	1
Old Litmus	37°	32	31	6	6	1	0
Dextrose agar		54	54	5	5	0	0
New Litmus	37°			1	0	0	0
Lactose agar				2	0	4	0
Old Litmus	37°	23	20	1	1	0	0
Lactose agar		29	28	0	0	0	0
New Bile	37°	5 neg.		5 neg.		5 neg.	
Old Bile	37°	5 neg.		5 neg.		5 neg.	

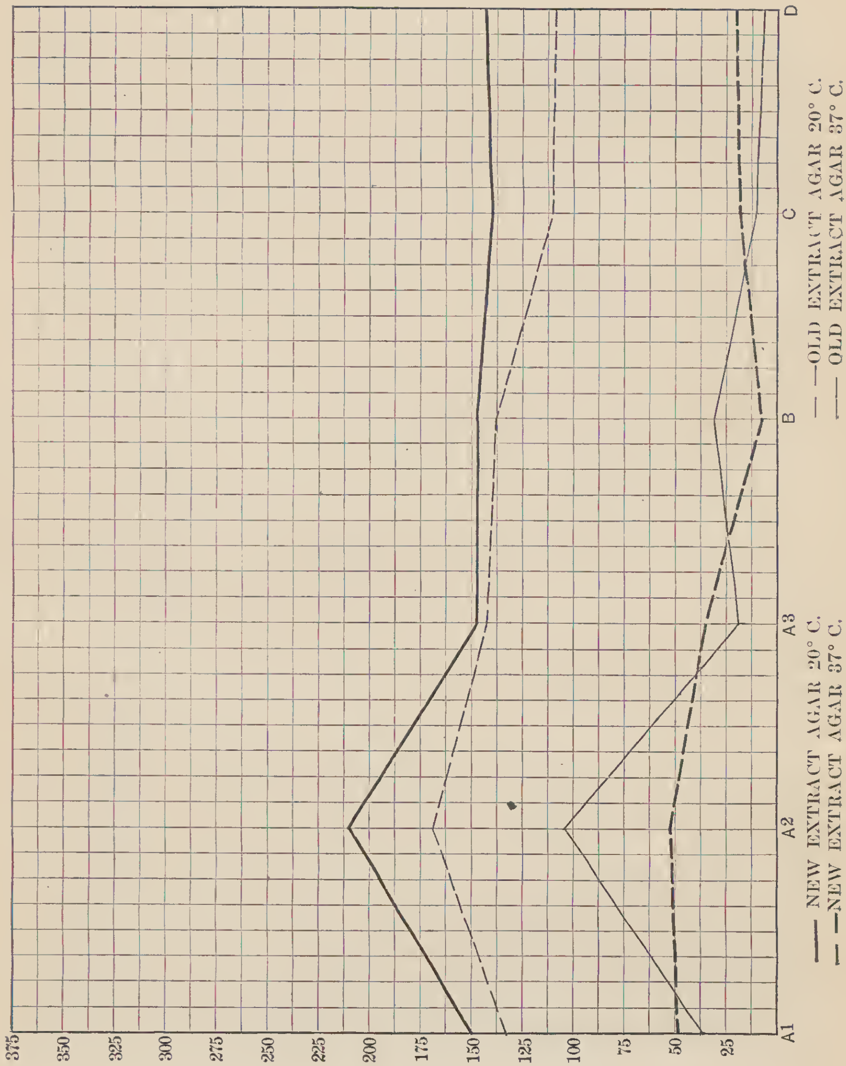
SAMPLE C.

Medium	Temp.	Bacteria per c.c. Dilution 1:10,000	Acid	Dilution 1:100,000	Acid	Dilution 1:1,000,000	Acid
New Meat agar	37°	51		spr.		1	
		66		spr.		0	
	20°	175		19		1	
		169		19		2	
Old Meat agar	37°	73		2		0	
		71		6		1	
	20°	74		10		3	
		66		7		0	
New Extract agar	37°	20		6		0	
		17		4		0	
	20°	134		16		2	
		149		17		2	
Old Extract agar	37°	12		0		spr.	
		9		3		2	
	20°	112		16		1	
		spr.		10		1	
New Casein agar	37°	5		4		3	
		7		4		1	
Old Casein agar	37°	10		1		4	
		5		6		2	
New Litmus	37°	spr.	..	1	1	1	0
Dextrose agar		22	22	1	0	0	0
Old Litmus	37°	23	23	1	0	0	0
Dextrose agar		20	20	0	0	spr.	..
New Litmus	37°	6	6	1	0	0	0
Lactose agar		8	4	0	0	0	0
Old Litmus	37°	12	10	0	0	0	0
Lactose agar		12	11	1	1	0	0
New Bile	37°	5 neg.		5 neg.		5 neg.	
Old Bile	37°	5 neg.		5 neg.		5 neg.	

SAMPLE D.

Medium	Temp.	Bacteria per c.c. Dilution 1:10,000	Acid	Dilution 1:100,000	Acid	Dilution 1:1,000,000	Acid
New Meat agar	37°	59		2		0	
		spr.		2		1	
	20°	199		19		1	
		172		27		2	
Old Meat agar	37°	spr.		6		1	
		82		spr.		2	
	20°	spr.		spr.		3	
		spr.		22		spr.	
New Extract agar	37°	19		1		4	
		20		7		0	
	20°	161		24		1	
		126		44		3	
Old Extract agar	37°	5		spr.		0	
		12		0		1	
	20°	99		10		2	
		122		11		1	
New Casein agar	37°	11		5		5	
		5		8		2	
Old Casein agar	37°	9		1		6	
		11		1		5	
New Litmus	37°	16	16	1	0	2	2
Dextrose agar		12	12	1	0	0	0
Old Litmus	37°	spr.	..	spr.	2	0	0
Dextrose agar		22	22	0	0	spr.	..
New Litmus	37°	spr.	..	3	2	0	..
Lactose agar		27	7	0	0	0	..
Old Litmus	37°	4	3	0	0	spr.	..
Lactose agar		13	9	1 spr.		0	0
New Bile	37°	5 neg.		5 neg.		5 neg.	
Old Bile	37°	5 neg.		5 neg.		5 neg.	


375
350
325
300
275
250
225
200
175
150
125
100
75
50
25
A1
A2
A3
B
C
D
NEW MEAT AGAR 20° C.
NEW MEAT AGAR 37° C.
OLD MEAT AGAR 37° C.
OLD MEAT AGAR 20° C.


375
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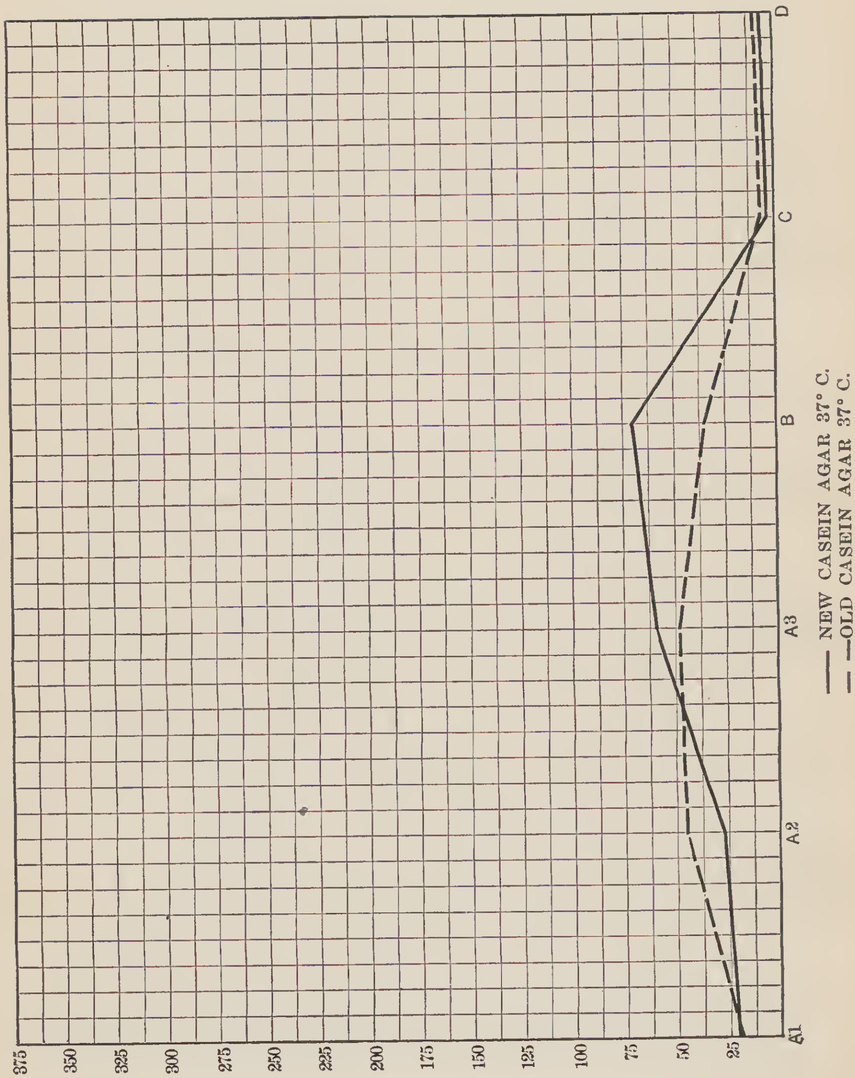
25
A1
A2
A3
B
C
D
NEW EXTRACT AGAR 20° C.
NEW EXTRACT AGAR 37° C.
OLD EXTRACT AGAR 20° C.
OLD EXTRACT AGAR 37° C.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A1
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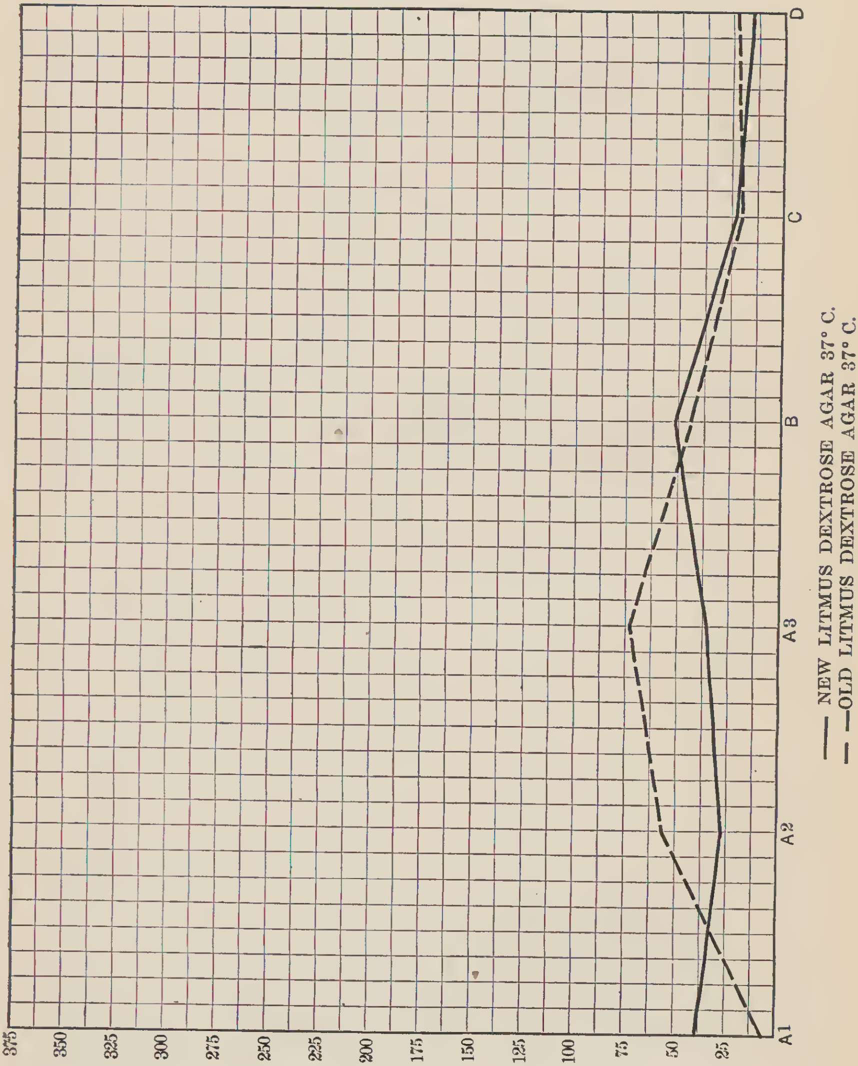
C
D
NEW CASEIN AGAR 37° C.
OLD CASEIN AGAR 37° C.


375
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200
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A1
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A3
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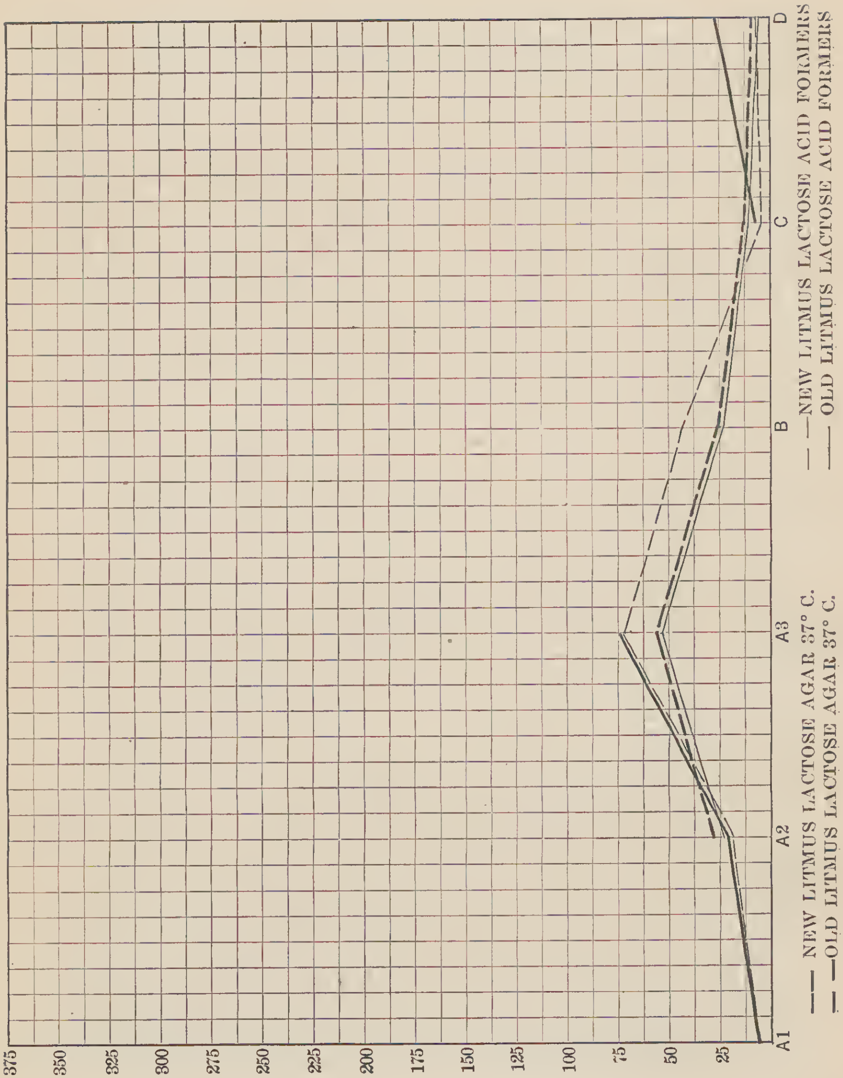
C
D
—— NEW LITMUS DEXTROSE AGAR 37° C.
— —OLD LITMUS DEXTROSE AGAR 37° C.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75
50
25
A1
A2
A3
B
C
D
NEW LITMUS LACTOSE AGAR 37° C.
OLD LITMUS LACTOSE AGAR 37° C.
NEW LITMUS LACTOSE ACID FORMERS
OLD LITMUS LACTOSE ACID FORMERS

EXAMINATION OF VANILLA ICE CREAM.

LOT 2, COMMERCIAL.

Determination of Effect of Different Media, Different Dilutions and Different Temperatures of Incubation on Numbers of Bacteria Found.

SAMPLE A1.

Medium	Temp.	Bacteria per c.c. Dilution 1 :10,000	Acid	Dilution 1 :100,000	Acid	Dilution 1 :1,000,000	Acid
New Meat agar	37°	1,360		233		13	
		2,148		253		spr.	
	20°	2,232		217		35	
		2,052		231		spr.	
Old Meat agar	37°	1,170		spr.		23	
		1,800		199		19	
	20°	1,500		231		32	
		1,560		219		28	
New Extract agar	37°	1,125		102		15	
		1,060		34		17	
	20°	1,058		191		27	
		1,416		132		33	
Old Extract agar	37°	1,788		90		18	
		900		108		19	
	20°	1,050		190		33	
		1,725		198		27	
New Casein agar	37°	890		91		16	
		1,530		70		14	
Old Casein agar	37°	1,530		130		16	
		1,524		126		spr.	
New Dextrose	37°	1,210	1,200	982	48	0	0
Lactose agar		960	957	90	85	0	0
Old Dextrose	37°	890	887	120	115	1	0
Lactose agar		460	457	106	96	spr.	0
New Litmus	37°	685	680	spr.	..	1	0
Lactose agar		760	757	92	88	spr.	..
Old Litmus	37°	930	925	89	83	0	0
Lactose agar		spr.		107	95	3	3
New Bile	37°	0		0		0	
		0		0		0	
		0		0		0	
Old Bile	37°	0		0		0	
		0		0		0	
		0		0		0	

SAMPLE A2.

Medium	Temp.	Bacteria per c.c. Dilution 1 :10,000	Acid	Dilution 1 :100,000	Acid	Dilution 1 :1,000,000	Acid
New Meat agar	37°	2,004		285		43	
		2,115		285		28	
	20°	spr.		249		38	
		1,764		258		29	
Old Meat agar	37°	1,890		265		30	
		2,240		261		31	
	20°	2,088		223		30	
		1,266		196		31	
New Extract agar	37°	1,212		..		..	
		1,308		151		17	
	20°	1,550		214		28	
		1,323		172		25	
Old Extract agar	37°	1,590		212		29	
		1,174		..		30	
	20°	1,608		235		44	
		1,968		225		34	
New Casein agar	37°	1,164		181		47	
		988		170		22	
Old Casein agar	37°	1,468		172		10	
		912		211		13	
New Litmus	37°	740	612	82	69	14	7
Dextrose agar		530	418	59	53	spr.	..
Old Litmus	37°	1,140	1,123	111	93	spr.	..
Dextrose agar		spr.	..	76	64	spr.	..
New Litmus	37°	1,000	988	107	102	24	24
Lactose agar		684	674	112	109	..	..
Old Litmus	37°	780	773	106	86	24	20
Lactose agar		800	780	121	118	7	2
New Bile	37°	0		0		0	
		0		0		0	
		0		0		0	
Old Bile	37°	0		0		0	
		0		0		0	
		0		0		0	

SAMPLE A3.

Medium	Temp.	Dilution 1 :10,000	Acid	Dilution 1 :100,000	Acid	Dilution 1 :1,000,000	Acid
		Bacteria per c.c.					
New Meat agar	37°	880		180		15	
		940		spr.		19	
	20°	1,827		247		45	
		1,974		248		38	
Old Meat agar	37°	1,020		192		8	
		1,020		190		6	
	20°	2,418		242		42	
		2,289		247		spr.	
New Extract agar	37°	820		63		8	
		960		72		10	
	20°	1,386		166		6	
		1,197		166		29	
Old Extract agar	37°	588		196		10	
		553		150		9	
	20°	990		168		19	
		1,056		208		26	
New Casein agar	37°	780		260		21	
		870		300		14	
Old Casein agar	37°	900		120		12	
		840		110		13	
New Dextrose	37°	spr.	839	155	115	6	2
Litmus agar		850		84	76	11	9
Old Dextrose	37°	1380	1336	128	114	spr.	..
Litmus agar		spr.	..	75	70	13	11
New Litmus	37°	910	896	146	129	18	16
Lactose agar		1210	1206	spr.*	..	spr.	..
Old Litmus	37°	900	890	115	104	18	18
Lactose agar		660	636	85	81	8	1
New Bile	37°	0		0		0	
		0		0		0	
		0		0		0	
Old Bile	37°	0		0		0	
		0		0		0	
		0		0		0	

* = alkaline.

SAMPLE B.

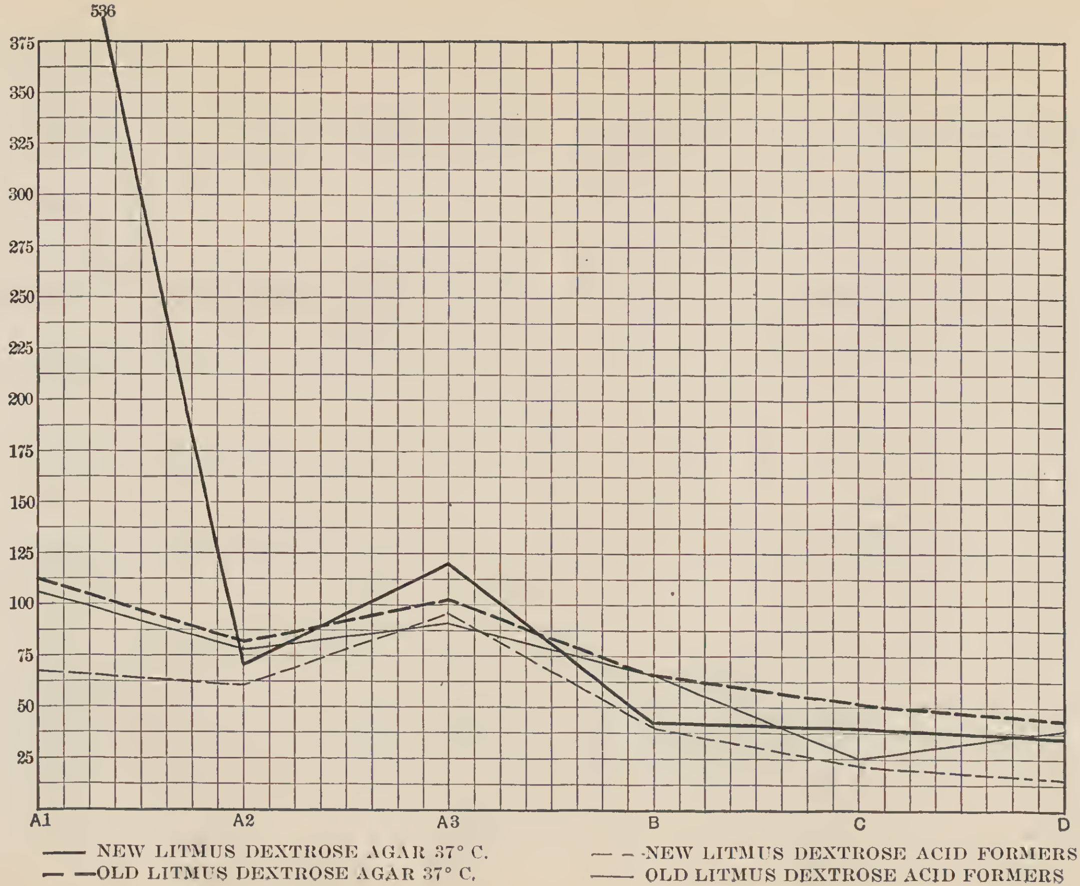
Medium	Temp.	Dilution 1 :10,000	Acid	Dilution 1 :100,000	Acid	Dilution 1 :1,000,000	Acid
		Bacteria per c.c.					
New Meat agar	37°	975		114		11	
		1,008		115		18	
	20°	1,890		205		22	
		1,915		205		30	
Old Meat agar	37°	spr.		134		18	
		1,400		128		spr.	
	20°	1,575		233		30	
		1,512		202		39	
New Extract agar	37°	888		122		15	
		828		133		24	
	20°	1,659		155		26	
		1,228		204		19	
Old Extract agar	37°	690		116		13	
		740		104		10	
	20°	693		174		23	
		1,449		186		16	
New Casein agar	37°	1,080		146		43	
		1,040		133		15	
Old Casein agar	37°	1,170		92		16	
		1,635		125		36	
New Litmus	37°	spr.	..	49	48	6	4
Dextrose agar		360	355	36	33	spr.	..
Old Litmus	37°	240	205	64	64	1	1
Dextrose agar		610	600	66	66	4	3
New Litmus	37°	700	690	63	58	11	10
Lactose agar		760	755	76	70	9	1
Old Litmus	37°	spr.	..	5	5	5	3
Lactose agar		490	485	62	57	10	9
New Bile	37°	0		0		0	
		0		0		0	
		0		0		0	
Old Bile	37°	0		0		0	
		0		0		0	
		0		0		0	

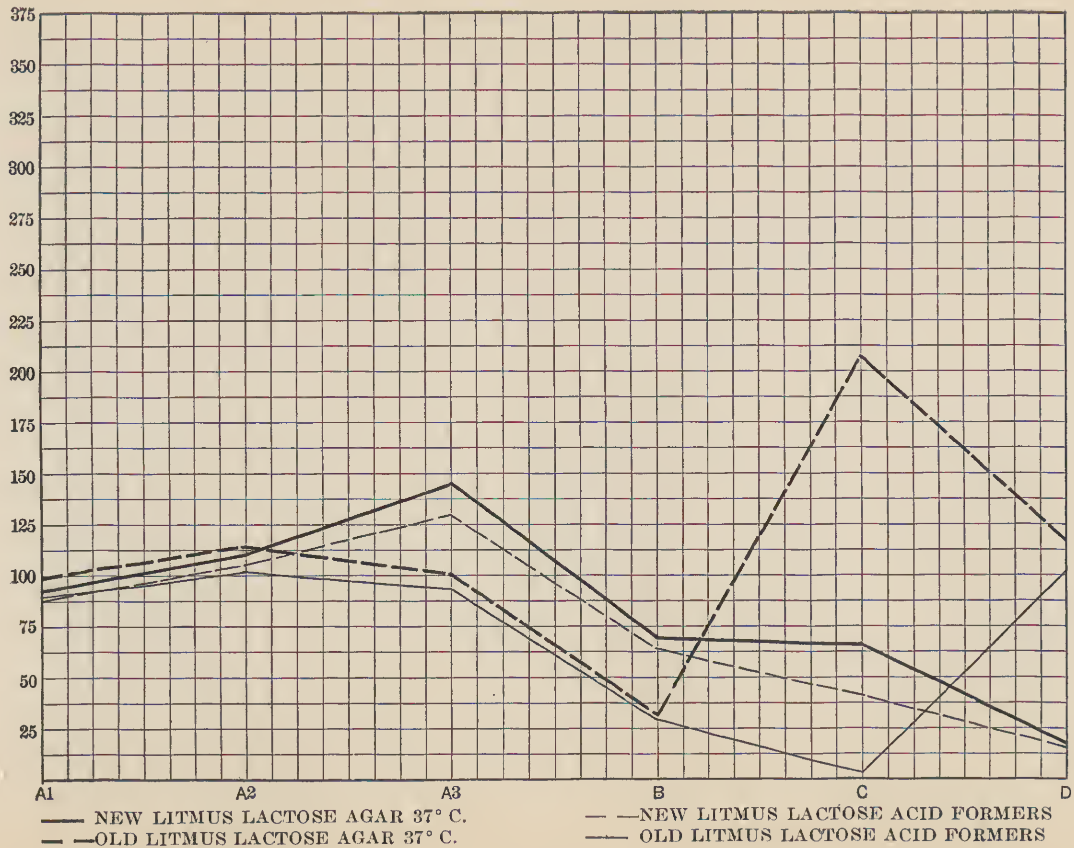
SAMPLE C.

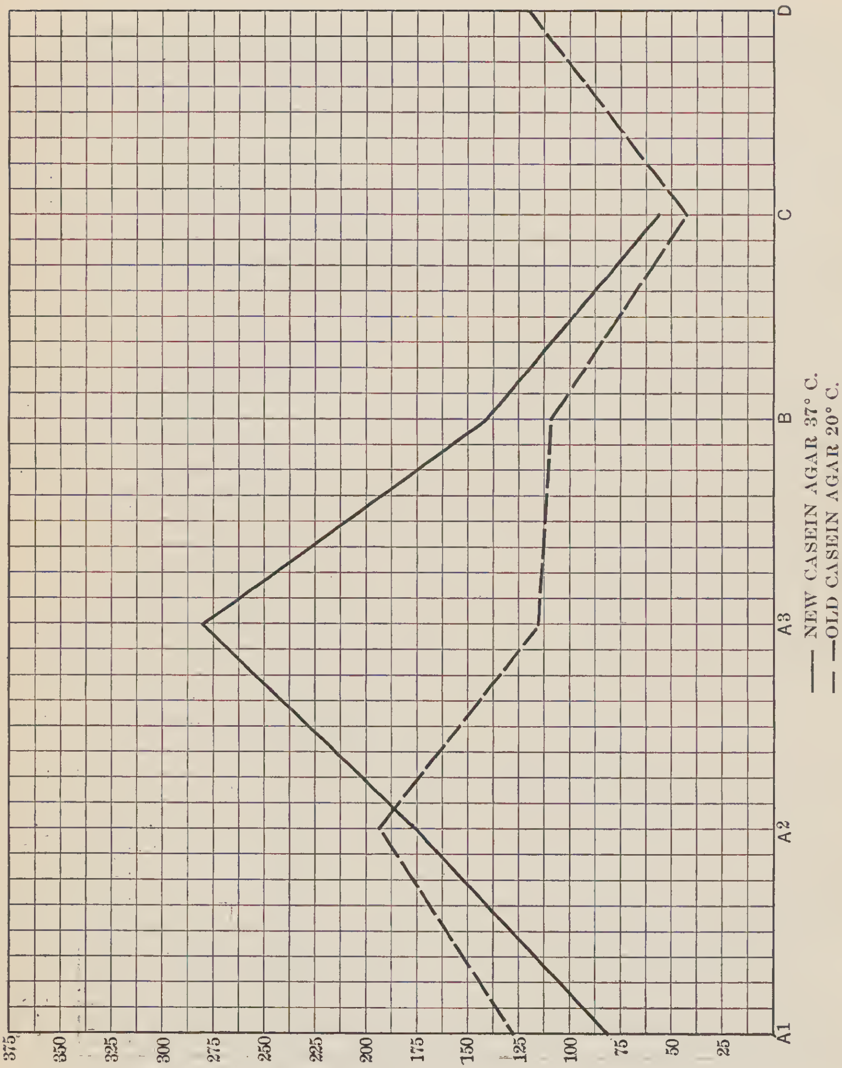
Medium	Temp.	Bacteria per c.c. Dilution 1 :10,000	Acid	Dilution 1 :100,000	Acid	Dilution 1 :1,000,000	Acid
New Meat agar	37°	500		75		3	
		spr.		spr.		11	
	20°	870		136		19	
		900		141		30	
Old Meat agar	37°	spr.		spr.		spr.	
		spr.		125		19	
	20°	1,150		150		23	
		660		spr.		23	
New Extract agar	37°	spr.		34		13	
		500		22		33	
	20°	1,032		156		20	
		1,134		132		19	
Old Extract agar	37°	202		32		213	
		480		22		7	
	20°	1,092		87		17	
		1,092		118		10	
New Casein agar	37°	220		54		28	
		560		61		21	
Old Casein agar	37°	340		49		6	
		400		34		8	
New Litmus	37°	427	417	spr.	..	4	1
Dextrose agar		160	5	40	21	1	1
Old Litmus	37°	spr.	..	34	28	alk.	spr.
Dextrose agar		340	330	68	23	2	1
New Litmus	37°	378	338	76	54	2	1
Lactose agar		spr.	..	56	30	1	0
Old Litmus	37°	452	240	271	6	15	11
Lactose agar		850	845	148	1	7	1
New Bile	37°	0		0		0	
		0		0		0	
		0		0		0	
Old Bile	37°	0		0		0	
		0		0		0	
		0		0		0	

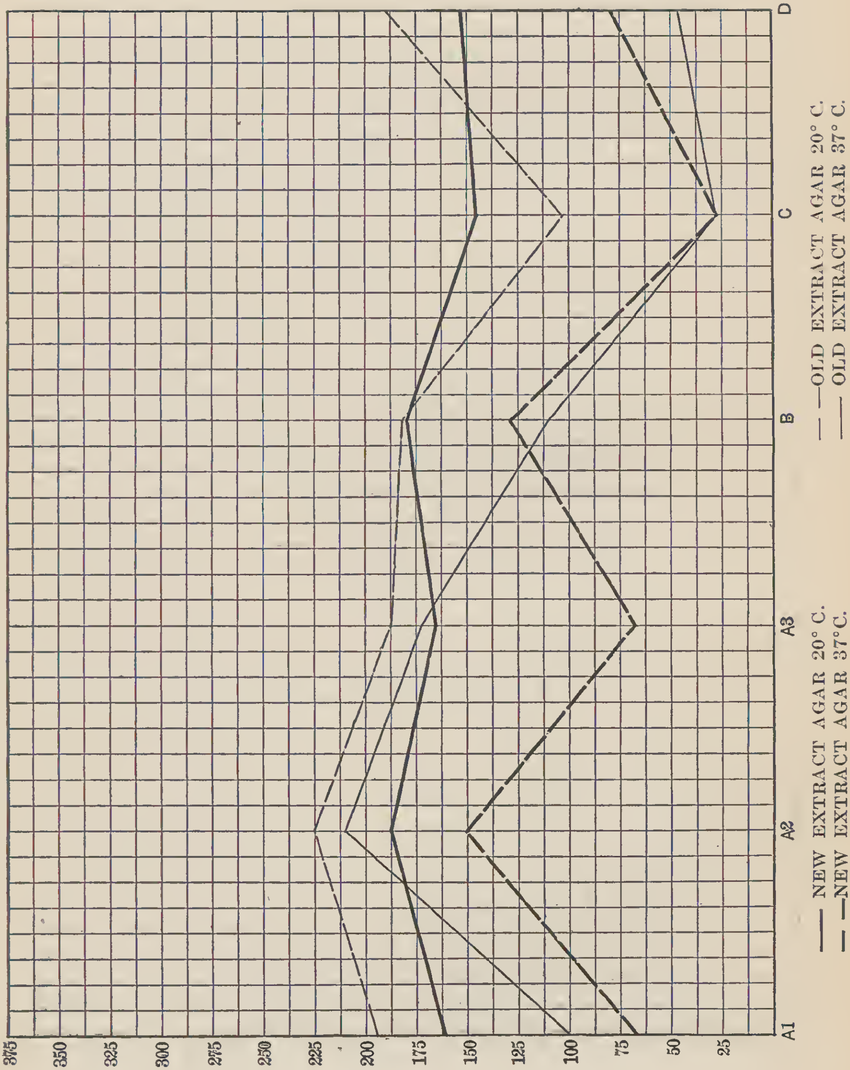
SAMPLE D.

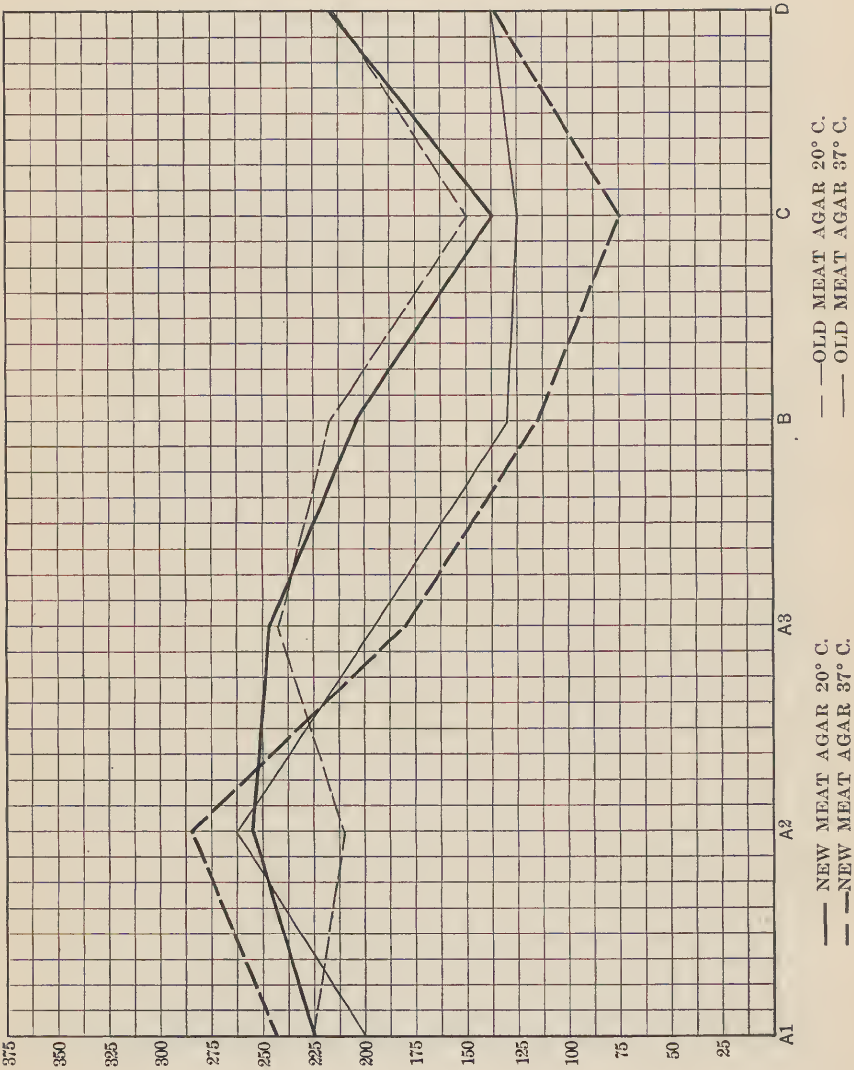
Medium	Temp.	Bacteria per c.c. Dilution 1 :10,000	Acid	Dilution 1 :100,000	Acid	Dilution 1 :1,000,000	Acid
New Meat agar	37°	spr.		144		spr.	
		670		126		29	
	20°	1.530		182		41	
		1,920		231		46	
Old Meat agar	37°	spr.		142		spr.	
		824		133		spr.	
	20°	1,356		215		29	
		1,692		spr.		31	
New Extract agar	37°	780		73		12	
		spr.		87		15	
	20°	1,092		160		34	
		1,548		146		38	
Old Extract agar	37°	209 spr.		47		17	
		240 spr.		48		10	
	20°	1,596		165		39	
		1,392		215		47	
New Casein agar	37°	not plated					
Old Casein agar	37°	1,392		103		26	
		696		139		21	
New Litmus	37°	1,300	1,293	34	14	4	0
Dextrose agar		1,080	1,073	spr.	..	6	4
Old Litmus	37°	1,180	1,170	35	34	spr.	..
Dextrose agar		970		47	42	5	1
New Litmus	37°	520	511	12	10	33	11
Lactose agar		1,270	1,265	23	22	8	1
Old Litmus	37°	650	640	spr.		spr.	
Lactose agar		spr.		117	103	9	6
New Bile	37°	0		0		0	
		0		0		0	
		0		0		0	
Old Bile	37°	0		0		0	
		0		0		0	
		0		0		0	


536
375
350
325
300
275
250
225
200
175
150
125
100
75
50
25
A1
A2
A3
B
C
D
NEW LITMUS DEXTROSE AGAR 37° C.
OLD LITMUS DEXTROSE AGAR 37° C.
NEW LITMUS DEXTROSE ACID FORMERS
OLD LITMUS DEXTROSE ACID FORMERS


375
350
325
300
275
250
225
200
175
150
125
100
75
50
25
A1
A2
A3
B
C
D
NEW LITMUS LACTOSE AGAR 37° C.
OLD LITMUS LACTOSE AGAR 37° C.
NEW LITMUS LACTOSE ACID FORMERS
OLD LITMUS LACTOSE ACID FORMERS


375
350
325
300
275
250
225
200
175
150
125
100
75
50
25
A1
A2
A3
B
C
D
—— NEW CASEIN AGAR 37° C.
— —OLD CASEIN AGAR 20° C.


375
350
325
300
275
250
225
200
175
150
125
100
75
50
25
A1
A2
A3
B
C
D
—— NEW EXTRACT AGAR 20° C.
— —NEW EXTRACT AGAR 37°C.
— —OLD EXTRACT AGAR 20° C.
—— OLD EXTRACT AGAR 37° C.


NEW MEAT AGAR 20° C.
NEW MEAT AGAR 37° C.
OLD MEAT AGAR 20° C.
OLD MEAT AGAR 37° C.
A1
A2
A3
B
C
D
375
350
325
300
275
250
225
200
175
150
125
100
75
50
25

Report on Ice Cream Examinations

P. G. HEINEMANN

REPORT ON ICE CREAM EXAMINATIONS MADE OCTOBER AND NOVEMBER, 1913.

P. G. Heinemann.

Methods.

The ice cream was bought in gallon cans. The samples were taken as follows:—The paper covering the ice cream was removed and a small layer of ice cream removed from the surface with a sterilized spoon. Four samples were then taken with sterile spoons and the ice cream transferred to sterile petri dishes and allowed to melt at room temperature. A sample was then taken by boring with a sterile glass tube one inch below the surface. The glass tubes used for this purpose were sterilized in test tubes, and after the sample had been taken were replacd in the tubes and the ice cream melted. The test tubes were stoppered with cotton. By using glass tubes as before one sample was taken two inches from surface, one three inches from surface. After these samples were taken the whole column of ice cream was dumped on a piece of sterilized filter paper. Hot water was run on the outside of the can until the ice cream would slip out. After this one sample was taken about three-quarters towards the bottom and two near the bottom. All these were melted in the respective test tubes. Finally two lots of about one half pint each were placed in sterilized beakers and allowed to melt. All samples excepting the two samples in the beakers were liquefied completely before samples were placed in dilution flasks. The two lots in the beakers were allowed to stand long enough to melt the ice cream so that one cubic centimeter could be removed.

The samples were designated as follows:

A1—Sample from beaker.
A2—Sample from beaker.
B1—Teaspoonful from surface.
B2—Teaspoonful from surface.
B3—Teaspoonful from surface.
B4—Teaspoonful from surface.
C1—Bored from two inches below surface.
C2—Bored from three inches below surface.
C3—Bored three-quarters way down.
C4—Bored from surface, about one inch down.
C5—Bored from bottom.
C6—Bored from bottom.

One cubic centimeter of each sample respectively was transferred to a flask containing 99 cc. sterile water. From this dilution one cubic centimeter was transferred to another flask containing 99 cc. sterile water. This makes a dilution of 1:10,000. From this dilution 10 cc. were transferred to a flask containing 90 cc. sterile water. All flasks were of course thoroughly shaken and all usual bacteriological manipulations carried on in proper shape. From the two dilutions 1:10,000 and 1:100,000 each one cc. was transferred to each of four petri dishes. To two of each set of four petri dishes one cc. of 1% litmus solution was added. This litmus was added with as much care as possible to avoid the mixing of the bacterial dilution with the litmus solution, the latter having some restraining effect upon bacteria. Mixing was effected after addition of the liquified agar. Two of each set of four petri dishes were used for plain agar, the other two for dextrose-litmus-agar. Plain agar is the

method used by the Committee on Standard Milk Analysis of the American Public Health Association and by the Government experts. Dextrose-litmus-agar favors the multiplication of lactic streptococci, the dextrose being an almost needed pabulum and the litmus facilitating the counting, since lactic streptococci form red colonies in litmus. After the agar had hardened the plates were incubated at 37 degrees C. for two days, and then the colonies counted. A separate count was made on all dextrose-litmus-agar plates to be able to get a relation of the acid forming bacteria to the others. This is especially important, since ice cream manufacturers often allow the cream to stand, which permits the multiplication of these acid organisms, which are entirely harmless. Colon bacilli would also multiply to some extent this way.

Tables 1 to 6 give the detailed counts. All plates of 1:10,000 and of 1:100,000 were counted, the dextrose-litmus-agar plates were counted twice in order to get the acid colony count. The dilution of 1:100,000 was not satisfactory, being too high and consequently giving high counts. The dilution 1:10,000 was satisfactory throughout. In the tables all these counts are given for the sake of completeness, but only the six counts of each sample were considered being of value. These six counts then were:—Two duplicate agar plates, which were averaged in a separate column. Two duplicate dextrose-litmus-agar plates, which were averaged in a separate column. The acid colonies on the two dextrose-litmus-agar plates, which were averaged in a separate column and then the percentage of acid colonies to the total estimated. The first six tables are too voluminous to give a definite idea of the results. In table 7 the results are condensed and show the total numbers of the bacteria found in a clear fashion.

TABLE 1

EXAMINATION OF ICE CREAM, LOT 1, CATERER, OCTOBER 26. PRICE $3.00.

EXPLANATION—A stands for plain agar, DLA for dextrose-litmus-agar. All figures should be multiplied by 1000.

Kind of Sample	No.	Dilution	Kind Med.	Plate No.	Colony count	Average count	Acid col. count	Average acid count	Per cent. acid col. count
A									
Each sample = ½ pint melted in sterile beaker.	1	10,000	A	1	250	210			
				2	170				
			DLA	1	450	395	200	175	45
				2	340		150		
		100,000	A	1	400	500			
				2	600				
			DLA	1	1,800	1,700	1,000	750	44
				2	1,600		500		
	2	10,000	A	1	800	725			
				2	650				
			DLA	1	1,200	1,150	500	450	39
				2	1,100		400		
		100,000	A	1	900	1,050			
				2	1,200				
			DLA	1	1,900	2,100	500	650	31
				2	2,300		800		
B									
Each sample = one teaspoonful taken from same place at surface.	1	10,000	A	1	400	410			
				2	420				
			DLA	1	480	480	200	200	42
				2	spreader				
		100,000	A	1	700	600			
				2	500				
			DLA	1	900	800	400	350	44
				2	700		300		
	2	10,000	A	1	700	750			
				2	800				
			DLA	1	1,300	1,500	300	350	23
				2	1,700		400		
		100,000	DLA	1	1,800	1,650	600	650	39
				2	1,500		700		
	3	10,000	A	1	180	195			
				2	210				
			DLA	1	450	415	50	45	11
				2	380		40		
		100,000	A	1	800	750			
				2	700				
			DLA	1	900	900	200	200	22
				2	spreader		200		
	4	10,000	A	1	120	105			
				2	90				
			DLA	1	320	340	150	150	44
				2	360		150		
		100,000	A	1	400	450			
				2	500				
			DLA	1	700	750	300	300	40
				2	800		300		

SAMPLES TAKEN FROM DIFFERENT PARTS:

C				1	270				
		10,000	A	2	330	300			
				1	820		300		
2 inches below surface.	1		DLA	2	760	790	300	300	38
				1	700				
		100,000	A	2	500	600			
				1	900		400		
			DLA	2	700	800	400	400	50
				1	650				
		10,000	A	2	840	745			
				1	1,600		200		
3 inches below surface.	2		DLA	2	1,300	1,450	300	250	17
				1	700				
		100,000	A	2	800	750			
				1	800		200		
			DLA	2	900	850	200	200	24
				1	960				
		10,000	A	2	780	870			
				1	1,800		400		
About ¾ ways down.	3		DLA	2	1,500	1,650	300	350	22
				1	1,900				
		100,000	A	2	2,500	2,200			
				1	3,200		1,200		
			DLA	2	2,100	2,650	300	750	28
				1	900				
		10,000	A	2	700	800			
				1	1,800		1,000		
Just below surface.	4		DLA	2	1,900	1,850	1,400	1,200	62
				1	spreaders				
		100,000	A	2	spreaders				
				1	spreaders				
			DLA	2	spreaders				
				1	360				
		10,000	A	2	380	370			
				1	750		200		
From bottom.	5		DLA	2	790	770	200	200	26
				1	900				
		100,000	A	2	500	700			
				1	spreaders				
			DLA	2	spreaders				
				1	1,040				
		10,000	A	2	900	870			
				1	1,160		280		
From bottom.	6		DLA	2	1,700	1,430	1,100	690	49
				1	1,200				
		100,000	A	2	1,900	1,550			
				1	3,200		800		
			DLA	2	1,200	2,200	1,200	1,000	45

TABLE 2

EXAMINATION OF LOT 2, COMMERCIAL ICE CREAM, OCTOBER 25, 1913. PRICE $1.25.

EXPLANATION—A stands for plain agar, DLA for dextrose-litmus-agar. All figures should be multiplied by 1000.

	Kind of Sample	No.	Dilution	Kind Med.	Plate No.	Colony count	Average count	Acid col. count	Average acid count	Per cent. acid col. count
A	Each sample = ½ pint melted in sterile beaker.	1	10,000	A	1	2,500				
					2	1,800	2,150			
				DLA	1	7,500		all		
					2	6,500	7,000	all	all	100
			100,000	A	1	3,800				
					2	2,400	3,100			
				DLA	1	9,200		8,000		
					2	8,400	8,800	5,000	6,500	74
		2	10,000	A	1	1,800				
					2	2,000	1,900			
				DLA	1	6,900		all		
					2	6,400	6,650	all	all	100
			100,000	A	1	1,100				
					2	1,400	1,250			
				DLA	1	2,900		1,000		
					2	3,400	3,150	1,200	1,100	35
B	Each sample = one teaspoonful taken from same place at surface.	1	10,000	A	1	1,900				
					2	2,400	2,150			
				DLA	1	9,500		all		
					2	8,500	9,000	all	all	100
			100,000	A	1	3,500				
					2	2,900	3,200			
				DLA	1	8,700		5,000		
					2	9,100	8,900	4,500	4,750	53
		2	10,000	A	1	2,000				
					2	3,600	2,800			
				DLA	1	3,750		all		
					2	2,500	3,100	1,500		75
			100,000	A	1	2,800				
					2	2,900	2,850			
				DLA	1	5,600		all		
					2	4,300	4,950	3,800		80
		3	10,000	A	1	2,900				
					2	2,500	2,700			
				DLA	1	9,200		all		
					2	8,500	8,850	all		100
			100,000	A	1	spreaders				
					2	1,100	1,100			
				DLA	1	spreaders				
					2	1,750	1,750	800		50
		4	10,000	A	1	2,800				
					2	2,500	2,650			
				DLA	1	7,600		5,700		
					2	8,500	8,050	all		75
			100,000	A	1	6,100				
					2	8,600	7,350			
				DLA	1	spreaders				
					2	19,000	19,000	8,500		50

SAMPLES TAKEN FROM DIFFERENT PARTS:

C				1	1,700				
		10,000	A	2	1,900	1,800			
				1	2,100		1,100		
2 inches below surface.	1		DLA	2	2,900	2,500	1,500	1,300	52
				1	3,800				
		100,000	A	2	2,800	3,300			
				1	9,300		all		
			DLA	2	8,100	8,700	all		100
				1	1,200				
		10,000	A	2	1,300	1,250			
				1	6,100		all		
3 inches below surface.	2		DLA	2	7,800	6,950	all		100
				1	2,200				
		100,000	A	2	2,700	2,450			
				1	9,500		5,000		
			DLA	2	7,600	8,050	5,800	5,400	68
				1	2,800				
		10,000	A	2	2,800	2,700			
				1	6,400		all		
About ¾ ways down.	3		DLA	2	9,800	8,100	all		100
				1	3,300				
		100,000	A	2	4,500	3,900			
				1	11,000		all		
			DLA	2	8,300	9,650	all		100
				1	2,300				
		10,000	A	2	3,400	2,850			
				1	8,400		4,500		
Just below surface.	4		DLA	2	9,500	8,950	all		75
				1	3,500				
		100,000	A	2	5,200	4,350			
				1	9,500		all		
			DLA	2	9,800	9,650	all		100
				1	5,200				
		10,000	A	2	3,900	4,550			
				1	9,200		all		
From bottom.	5		DLA	2	11,200	10,200	all		100
				1	8,000				
		100,000	A	2	13,000	10,500			
				1	14,000		12,000		
			DLA	2	15,000	14,500	8,000	10,000	69
				1	2,600				
		10,000	A	2	2,400	2,500			
				1	6,900		all		
From bottom.	6		DLA	2	4,200	5,500	all		100
				1	2,200				
		100,000	A	2	3,600	2,900			
				1	7,500		all		
			DLA	2	8,900	8,200	all		100

TABLE 3

EXAMINATIONS OF LOT 3, COMMERCIAL ICE CREAM, OCTOBER 24, 1913. PRICE $1.70.

EXPLANATION—A stands for plain agar, DLA for dextrose-litmus-agar. All figures should be multiplied by 1000.

Kind of Sample	No.	Dilution	Kind Med.	Plate No.	Colony count	Average count	Acid col. count	Average acid count	Per cent. acid col. count
A				1	240				
		10,000	A	2	300	270			
				1	700		100		
Each sample = ½ pint melted in sterile beaker.	1		DLA	2	650	675	100	100	15
				1	300				
		100,000	A	2	200	250			
				1	400		none		
			DLA	2	500	450	none	none	0
				1	270				
		10,000	A	2	300	285			
				1	420		310		
	2		DLA	2	480	450	360	335	75
				1	100				
		100,000	A	2	200	150			
				1	200		200		
			DLA	2	200	200	200	200	100
B				1	190				
		10,000	A	2	160	175			
				1	600		300		
Each sample = one teaspoonful taken from same place at surface.	1		DLA	2	500	550	250	275	50
				1	400				
		100,000	A	2	200	300			
				1	400		200		
			DLA	2	400	400	200	200	50
				1	410				
		10,000	A	2	370	390			
				1	2,240		1,800		
	2		DLA	2	3,250	2,700	2,650	2,220	82
				1	720				
		100,000	A	2	580	650			
				1	1,180		all		
			DLA	2	2,300	1,740	all	all	100
				1	290				
		10,000	A	2	250	270			
				1	460		200		
	3		DLA	2	370	415	120	160	39
				1	600				
		100,000	A	2	500	550			
				1	1,100		550		
			DLA	2	1,800	1,450	400	475	33
				1	800				
		10,000	A	2	900	850			
				1	3,900		2,200		
	4		DLA	2	3,100	3,500	1,900	2,050	59
				1	200				
		100,000	A	2	300	250			
				1	400		300		
			DLA	2	700	550	400	350	64

C

SAMPLES TAKEN FROM DIFFERENT PARTS:

2 inches below surface.	1	10,000	A	1	160				
				2	120	140			
			DLA	1	650		500		
				2	510	580	450	475	93
		100,000	A	1	200				
				2	100	150			
			DLA	1	400		200		
				2	300	350	100	150	43
3 inches below surface.	2	10,000	A	1	450				
				2	400	425			
			DLA	1	2,500		1,900		
				2	2,900	2,700	2,500	2,200	81
		100,000	A	1	1,100				
				2	1,900	1,500			
			DLA	1	1,200		800		
				2	1,600	1,400	600	700	50
About ¾ way down.	3	10,000	A	1	160				
				2	180	170			
			DLA	1	700		600		
				2	900	800	400	500	62
		100,000	A	1	300				
				2	400	350			
			DLA	1	1,100		700		
				2	800	850	400	550	65
Just below surface.	4	10,000	A	1	800				
				2	900	850			
			DLA	1	2,200		800		
				2	1,800	2,000	1,200	1,000	50
		100,000	A	1	2,000				
				2	2,000	2,000			
			DLA	1	4,500		2,100		
				2	3,900	4,200	2,200	2,150	50
From bottom.	5	10,000	A	1	700				
				2	500	600			
			DLA	1	850		700		
				2	650	750	620	660	88
		100,000	A	1	600				
				2	800	700			
			DLA	1	900		300		
				2	1,200	1,050	400	350	33
From bottom.	6	10,000	A	1	390				
				2	360	370			
			DLA	1	750		700		
				2	800	775	750	725	93
		100,000	A	1	1,100				
				2	800	850			
			DLA	1	1,600		1,500		
				2	1,400	1,500	1,200	1,350	90

TABLE 4

EXAMINATIONS OF LOT 4, COMMERCIAL VANILLA ICE CREAM, OCTOBER 23, 1913. PRICE $1.25.

EXPLANATION—A stands for plain agar, DLA for dextrose-litmus-agar. All figures should be multiplied by 1000.

Kind of Sample	No.	Dilution	Kind Med.	Plate No.	Colony count	Average count	Acid col. count	Average acid count	Per cent. acid col. count
A									
		10,000	A	1	9,920				
				2	7,800	8,850			
	1		DLA	1	22,000		all		
				2	18,000	20,000	all	all	100
		100,000	A	1	12,000				
Each sample = ½ pint melted in sterile beaker.				2	8,000	10,000			
			DLA	1	21,000		all		
				2	18,000	19,500	all	all	100
Sample taken when enough melted.		10,000	A	1	5,840				
				2	7,120	6,480			
	2		DLA	1	16,000		all		
				2	23,000	19,500	all	all	100
		100,000	A	1	6,000				
				2	8,000	7,000			
				1	25,000		17,000		
				2	28,500	26,500	25,000	21,000	79
B									
		10,000	A	1	10,900				
				2	12,000	11,500			
Each sample = one teaspoonful taken from same place at surface.	1		DLA	1	33,000		all		
				2	33,600	33,300	all	all	100
		100,000	A	1	11,000				
				2	11,000	11,000			
				1	39,000		all		
				2	50,000	44,500	all	all	100
		10,000	A	1	9,200				
				2	13,000	11,000			
	2		DLA	1	24,000		all	all	100
				2	26,000	25,000	all		
		100,000	A	1	14,000				
				2	10,000	12,000			
			DLA	1	40,000		all		
				2	45,000	42,500	all	all	100
		10,000	A	1	43,000				
				2	46,400	44,700			
	3		DLA	1	48,000		all	all	100
				2	46,000	47,000	all		
		100,000	A	1	45,000				
				2	48,000	46,500			
			DLA	1	58,000		all		
				2	56,000	57,000	all	all	100
		10,000	A	1	41,000				
				2	42,000	41,500			
			DLA	1	59,000		all		
	4		DLA	1	59,000	55,000	all	all	100
		100,000	A	1	60,000				
				2	50,000	55,000			
			DLA	1	55,000		all		
				2	65,000	60,000	all	all	100

C

SAMPLES TAKEN FROM DIFFERENT PARTS:

2 inches below surface.	1	10,000	A	1	6,800				
				2	6,200	6,500			
			DLA	1	29,000		all		
				2	22,000	25,500	all	all	100
		100,000	A	1	21,000				
				2	11,500	16,200			
			DLA	1	38,000		all		
				2	36,000	37,000	all	all	100
3 inches below surface.	2	10,000	A	1	7,500				
				2	6,500	7,000			
			DLA	1	31,000		all		
				2	29,000	30,000	all	all	100
		100,000	A	1	17,000				
				2	14,000	15,500			
			DLA	1	48,000		all		
				2	43,000	45,500	all	all	100
About ¾ way down.	3	10,000	A	1	8,500				
				2	9,700	9,100			
			DLA	1	20,400		all		
				2	27,200	23,800	all	all	100
		100,000	A	1	11,000				
				2	9,000	10,000			
			DLA	1	35,000		all		
				2	28,800	31,900	all	all	100
Just below surface.	4	10,000	A	1	9,900				
				2	8,600	9,200			
			DLA	1	40,400		all		
				2	35,000	37,500	all	all	100
		100,000	A	1	18,500				
				2	17,500	18,000			
			DLA	1	58,700		all		
				2	55,200	56,900	all	all	100
From bottom.	5	10,000	A	1	8,500				
				2	8,800	8,600			
			DLA	1	24,000		all		
				2	20,500	22,500	all	all	100
		100,000	A	1	11,000				
				2	12,000	10,500			
			DLA	1	29,800		all		
				2	32,000	29,900	all	all	100
From bottom.	6	10,000	A	1	5,900				
				2	7,300	6,600			
			DLA	1	18,400		all		
				2	17,500	17,900	all	all	100
		100,000	A	1	7,500				
				2	6,500	7,000			
			DLA	1	19,000		all		
				2	16,000	17,500	all	all	100

TABLE 5

EXAMINATION OF LOT 5, COMMERCIAL STRAWBERRY ICE CREAM, NOVEMBER 7, 1913. PRICE $1.25.

EXPLANATION—A stands for plain agar, DLA for dextrose-litmus-agar. All figures should be multiplied by 1,000.

Kind of Sample	No.	Dilution	Kind Med.	Plate No.	Colony count	Average count	Acid col. count	Average acid count	Per cent. acid col. count
A				1	950				
		10,000	A	2	750	850			
				1	980		900		
	1		DLA	2	spreaders	980	900	900	80
				1	700				
		100,000	A	2	800	750			
Each sample = ½ pint melted in sterile beaker.				1	900		300		
			DLA	2	1,100	1,000	900	600	60
				1	650				
		10,000	A	2	720	685			
				1	850		650		
	2		DLA	2	900	875	800	725	90
				1	500				
		100,000	A	2	700	600			
				1	1,000		800		
			DLA	2	spreaders	1,000		800	80
B				1	800				
		10,000	A	2	650	725			
				1	850		750		
	1		DLA	2	950	900	500	625	68
				1	900				
		100,000	A	2	800	850			
				1	800		700		
			DLA	2	900	800	700	700	88
				1	880				
		10,000	A	2	750	815			
				1	1,200		400		
	2		DLA	2	900	1,050	700	550	54
				1	800				
		100,000	A	2	1,100	950			
Each sample = one teaspoonful from same place at surface.				1	1,300		1,300		
			DLA	2	spreaders			1,300	100
				1	1,300				
		10,000	A	2	1,100	1,200			
				1	1,100		1,000		
	3		DLA	2	spreaders	1,100		1,000	91
				1	1,500				
		100,000	A	2	1,100	1,300			
				1	1,800		1,500		
			DLA	2	spreaders			1,500	83
				1	950				
		10,000	A	2	800	875			
				1	900		800		
	4		DLA	2	950	925	700	750	75
				1	1,100				
		100,000	A	2	1,300	1,200			
				1	1,500		1,200		
			DLA	2	1,800	1,650	1,500	1,350	82

C

SAMPLES TAKEN FROM DIFFERENT PARTS:

		10,000	A	1	950				
				2	1,100	1,025			
			DLA	1	1,400		1,200		
2 inches below surface.	1			2	1,200	1,300	1,000	1,100	85
		100,000	A	1	1,300				
				2	900	1,100			
			DLA	1	spreaders				
				2	1,600	1,600	1,600	1,600	100
		10,000	A	1	900				
				2	1,200	1,050			
			DLA	1	920		900		
3 inches below surface.	2			2	1,100	1,020	1,000	950	95
		100,000	A	1	1,200				
				2	1,800	1,500			
			DLA	1	1,800		1,500		
				2	2,000	1,900	1,500	1,500	79
		10,000	A	1	820				
				2	930	875			
			DLA	1	1,100		1,100		
About ¾ way down.	3			2	1,200	1,150	1,000	1,050	90
		100,000	A	1	700				
				2	900	800			
			DLA	1	1,200		800		
				2	1,500	1,350	1,200	1,000	74
		10,000	A	1	1,200				
				2	1,050	1,125			
			DLA	1	1,300		1,300		
Just below surface.	4			2	1,600	1,450	1,200	1,250	86
		100,000	A	1	1,200				
				2	1,300	1,250			
			DLA	1	1,600		1,200		
				2	1,400	1,500	1,100	1,150	77
		10,000	A	1	600				
				2	550	575			
			DLA	1	800		700		
From bottom.	5			2	850	825	700	700	85
		100,000	A	1	1,200				
				2	1,400	1,300			
			DLA	1	spreaders				
				2	1,500	1,500	1,500	1,500	100
		10,000	A	1	700				
				2	850	775			
			DLA	1	1,600		1,500		
From bottom.	6			2	1,050	1,325	900	1,200	90
		100,000	A	1	1,300				
				2	1,400	1,350			
			DLA	1	1,400		1,400		
				2	1,200	1,300	1,000	1,200	92

TABLE 6

EXAMINATION OF VANILLA ICE CREAM, LOT 6, COMMERCIAL, NOVEMBER 9, 1913. PRICE $1.25.

EXPLANATION—A stands for plain agar, DLA for dextrose-litmus-agar. All figures should be multiplied by 1,000.

Kind of Sample	No.	Dilution	Kind Med.	Plate No.	Colony count	Average count	Acid col. count	Average acid count	Per cent. acid col. count
A									
Each sample = ½ pint melted in sterile beaker.	1	10,000	A	1	460				
				2	480	470			
			DLA	1	700		700		
				2	660	680	660	680	100
		100,000	A	1	500				
				2	1,100	800			
			DLA	1	700		400		
				2	1,200	950	700	550	45
	2	10,000	A	1	1,000				
				2	2,400	1,700			
			DLA	1	4,200		4,200		
				2	3,600	3,900	3,000	3,600	95
		100,000	A	1	5,000				
				2	7,000	6,000			
			DLA	1	10,000		8,000		
				2	spreaders	10,000	8,000	8,000	80
B									
Each sample = one teaspoonful from same place at surface.	1	10,000	A	1	780				
				2	900	840			
			DLA	1	920		920		
				2	960	940	960	940	100
		100,000	A	1	600				
				2	900	750			
			DLA	1	1,000		800		
				2	1,700	1,350	1,700	1,250	93
	2	10,000	A	1	540				
				2	840	690			
			DLA	1	1,400		1,400		
				2	1,020	1,210	1,020	1,210	100
		100,000	A	1	400				
				2	700	550			
			DLA	1	1,100		1,100		
				2	1,900	1,500	1,900	1,500	100
	3	10,000	A	1	700				
				2	800	750			
			DLA	1	900		800		
				2	740	820	500	650	80
		100,000	A	1	500				
				2	600	650			
			DLA	1	800		500		
				2	900	750	400	450	60
	4	10,000	A	1	480				
				2	540	510			
			DLA	1	1,200		1,200		
				2	1,100	1,150	900	1,050	91
		100,000	A	1	500				
				2	600	550			
			DLA	1	700		400		
				2	800	750	600	500	67

C

SAMPLES TAKEN FROM DIFFERENT PARTS:

				1	350				
		10,000	A	2	390	370			
				1	420		350		
2 inches below surface.	1		DLA	2	spreaders	420		350	83
				1	300				
		100,000	A	2	400	350			
				1	700		500		
			DLA	2	600	650	400	450	70
				1	350				
		10,000	A	2	400	375			
				1	520		520		
3 inches below surface.	2		DLA	2	560	540	560	540	100
				1	300				
		100,000	A	2	200	150			
				1	400		300		
			DLA	2	500	450	500	400	89
				1	200				
		10,000	A	2	380	290			
				1	410		410		
About ¾ way down.	3		DLA	2	370	390	370	390	100
				1	700				
		100,000	A	2	600	650			
				1	800		800		
			DLA	2	spreaders	800	800	800	100
				1	210				
		10,000	A	2	170	190			
				1	350		350		
Just below surface.	4		DLA	2	280	315	280	315	100
				1	200				
		100,000	A	2	200	200			
				1	500		200		
			DLA	2	800	650	600	400	60
				1	450				
		10,000	A	2	380	415			
				1	740		740		
From bottom.	5		DLA	2	560	650	500	620	95
				1	600				
		100,000	A	2	700	650			
				1	1,200		800		
			DLA	2	800	1,000	500	650	65
				1	300				
		10,000	A	2	400	350			
				1	900		900		
From bottom.	6		DLA	2	800	850	800	850	100
				1	800				
		100,000	A	2	600	700			
				1	1,200		1,200		
			DLA	2	800	1,000	800	1,000	100

A

TABLE 7

CONDENSED TABLE OF RESULTS.

EXPLANATION—A stands for plain agar, DLA for dextrose-litmus-agar. All figures should be multiplied by 1,000.

			ICE CREAM SAMPLE: 1		2		3		4		5		6	
Sample		Medium	Count	% acid	Count	% acid	Count	% acid	Count	% acid	Count	% acid	Count	% acid
		A	210		2,150		270		8,850		850		470	
	1	DLA	395	45	7,000	100	675	15	20,000	100	980	89	680	100
A		A	725		1,900		285		6,480		685		1,700	
	2	DLA	1,150	39	6,650	100	450	75	19,500	100	875	90	3,900	95
		A	410		2,150		175		11,500		725		840	
	1	DLA	480	42	9,000	100	550	50	33,300	100	900	68	940	100
		A	750		2,800		390		11,000		815		690	
B	2	DLA	1,500	23	3,100	75	2,700	82	25,000	100	1,050	54	1,210	100
		A	195		2,700		270		44,700		1,200		750	
	3	DLA	415	11	8,850	100	415	39	47,000	100	1,000	91	820	80
		A	105		2,800		850		41,500		875		510	
	4	DLA	340	44	8.850	100	3,500	59	55,000	100	925	75	1,150	91
		A	300		1.800		140		6,500		1,025		370	
	1	DLA	790	38	2,500	52	580	93	25,500	100	1,300	85	420	83
		A	745		1.250		425		7,000		1.050		375	
	2	DLA	1,450	17	6,950	100	2,700	81	30,000	100	1,020	95	540	100
		A	870		2,700		170		9,100		875		290	
C	3	DLA	1,650	22	8,100	100	800	62	23,800	100	1,150	90	390	100
		A	800		2.850		850		9,200		1,125		190	
	4	DLA	1,850	62	8,950	75	2,000	50	37,500	100	1,450	86	315	100
		A	370		4,550		600		8,600		575		415	
	5	DLA	770	26	10,200	100	750	88	22,500	100	825	85	650	95
		A	870		2,500		370		6,600		775		350	
	6	DLA	1,430	49	5,500	100	775	93	17,900	100	1,325	90	850	100

Sample A 1 and sample A 2 represent the amount liquefied ice cream from one-half pint in a sterile beaker. One cc was taken as soon as a sufficient amount was melted.

Sample B—All four samples were taken with sterile teaspoons from the same place at the surface, after scraping a thin layer off.

Sample C—All "C" samples were taken with sterile glass tubes by boring into the ice cream.
1 was taken 2 inches below surface.
2 was taken 3 inches below surface.
3 was taken about three-quarters down from surface.
4 was the first boring from surface.
5 and 6 were taken from the bottom.

B AVERAGES:

Sample	Plain agar	Dextrose-litmus-agar	Per cent. acid colonies
1	530,000	1,020,000	35
2	2,510,000	7,140,000	92
3	400,000	1,325,000	66
4	14,250,000	29,810,000	100
5	881,000	1,106,000	83
6	579,000	989,000	95

C AVERAGES ACCORDING TO PART OF GALLON SAMPLE WAS TAKEN.

All figures to be multiplied by 1,000.

		ICE CREAM SAMPLE NUMBER:							
Sample	Med.	1	2	3	4	5	6	Average	Ratio
Melted in beaker.	A	468	2,025	277	7,665	767	2,170	2,227	11
	DLA	773	6,825	563	19,750	927	2,290	5,188	26
From surface.	A	365	2,575	421	27,175	904	697	5,356	27
	DLA	684	7,250	1,791	40,750	994	1,030	8,750	44
2 in. below surface.	A	300	1,800	140	6,500	1,025	370	1,689	8
	DLA	790	2,500	580	25,500	1,300	420	5,682	28
3 in. below surface.	A	745	1,250	425	7,000	1,050	375	1,807	9
	DLA	1,450	6,950	2,700	30,000	1,020	540	7,110	35
¾ th ways down.	A	870	2,700	170	9,100	875	290	2,334	12
	DLA	1,650	8,100	800	23,800	1,150	390	5,982	30
Just below surface.	A	800	2,850	850	9,200	1,125	190	2,502	12
	DLA	1,850	8,950	2,000	37,500	1,450	315	8,677	43
From bottom.	A	620	3,525	485	7,600	675	500	2,234	11
	DLA	1,100	7,750	762	21,100	1,100	675	5,277	26

TABLE 7A

RELATION OF NUMBERS OF LACTIC ACID STREPTOCOCCI TO AMOUNT OF ACID PRODUCED UNDER DIFFERENT TEMPERATURE CONDITIONS.

Per cent. acid is expressed in lactic acid. All numbers should be multiplied by 1,000.

Kind of Milk	Temp.	Incub. period	Colony Count: Plain agar	Colony Count: Dext.-lit.-agar	Per cent. acid col.	Per cent. Lact. acid	Remarks
Raw Milk	37° C	fresh	36	160	94	0.12	
		1 day	238,000	359,000	100	0.59	Sour, curd and whey.
		2	272,000	432,000	100	0.63	Sour, curd and whey.
		3	289,000	361,000	100	0.67	Sour, curd and whey.
		4	268,000	401,000	100	0.69	Sour, curd and whey.
		5		spreaders		1.24	Sour, curd and whey.
		6	138,000	160,000	100	1.52	Sour, curd and whey.
Raw Milk	21° C	fresh	36	160	94	0.12	
		1 day	156,000	185,000	100	0.26	Turning.
		2	252,000	270,000	100	0.54	Sour, curd and whey.
		3	480,000	820,000	100	0.68	Sour, curd and whey.
		4	350,000	730,000	100	0.74	Sour, curd and whey.
		5		spreaders		0.76	Sour, curd and whey.
		6	117,000	195,000	49	0.78	Sour, curd and whey.
Raw Milk	7° C	fresh	36	160	94	0.12	
		1 day	54	170	100	0.12	No change.
		2	55	256	100	0.12	No change.
		3	150	190	100	0.12	No change.
		4	4,100	10,100	100	0.14	No change.
		5	6,500	7,400	100	0.15	No change.
		6	15,000	31,000	45	0.16	Bitter taste.
		7	22,000	29,000	47	0.20	Bitter taste.
		8	31,000	44,000	45	0.24	Very bitter.
		9	47,000	52,000	55	0.24	Very bitter.
Raw Cream	37° C	fresh	28	175	100	0.12	
		1 day	485,000	1,285,000	100	0.71	Sour, homog. curd.
		2	595,000	1,300,000	100	0.73	Sour, homog. curd.
		3	44,000	65,000	22	1.02	Sour, thick.
		4	54,000	68,000	21	1.18	Sour, very thick.
		5		spreaders		1.40	Sour, very thick.
		6	66,000	85,000	00	1.61	Sour, bitter, thick.
Raw Cream	21° C	fresh	28	175	100	0.12	
		1 day	55,000	156,000	100	0.26	Sweet, homogeneous.
		2	135,000	350,000	100	0.65	Sour, thick.
		3	210,000	390,000	85	0.67	Sour, thick.
		4	270,000	380,000	80	0.69	Sour, thick.
		5	250,000	410,000	75	0.67	Sour, thick.
		6	310,000	460,000	80	0.72	Sour, thick.

Raw Cream	7° C	fresh	28	175	100	0.12	
		1 day	38	214	100	0.12	No change.
		2	290	1,050	100	0.12	No change.
		3	4,100	7,300	71	0.13	No change.
		4	8,500	11,300	91	0.15	No change.
		5	26,500	29,000	100	0.16	No change.
		6	36,000	53,000	39	0.18	No change.
		7	29,000	31,000	100	0.26	No change.
		8	37,000	42,000	000	0.30	Turning.
		9	85,000	120,000	000	0.34	Bitter.
Sterile Milk	37° C	fresh	9	26	100	0.12	
		1 day	199,200	220,000	100	0.51	Sour, curd.
		2	250,000	240,000	100	0.63	Sour, curd.
		3	213,000	317,000	100	0.66	Sour, curd.
		4		spreaders		0.69	Sour, curd.
		5	224,000	339,000	100	0.99	Sour, curd.
		6	219,900	433,100	100	1.28	Sour, curd.
Sterile Milk	21° C	fresh	9	26	100	0.12	
		1 day	162,000	202,000	100	0.39	Sour, light curd.
		2	288,000	353,000	100	0.52	Sour, curd.
		3	310,000	680,000	100	0.66	Sour, curd.
		4	380,000	640,000	100	0.72	Sour, curd.
		5		spreaders		0.73	Sour, curd.
		6	415,000	760,000	100	0.76	Sour, curd.
Sterile Milk	7° C	fresh	9	26	100	0.12	
		1 day	1,700	2,500	100	0.16	No change.
		2	16,000	21,000	100	0.17	No change.
		3	19,000	32,000	100	0.18	No change.
		4	25,000	36,000	100	0.19	No change.
		5	32,000	59,000	100	0.20	No change.
		6	35,000	43,000	100	0.20	No change.
		7	72,000	117,000	100	0.23	No change.
		8	78,000	160,000	100	0.27	No change.

RELATION OF NUMBERS OF LACTIC ACID STREPTOCOCCI TO AMOUNT OF ACID PRODUCED UNDER DIFFERENT TEMPERATURE CONDITIONS.

The numbers in the table include all bacteria in the samples of raw milk and raw cream, excepting under the column "acid colonies." In sterlized milk the numbers are streptococci only. All flasks were inoculated with a pure culture of lactic streptococci. Counts were made immediately after inoculation and after that at intervals of 24 hours. One set of flasks was kept at 7° C., another set at 21° C. and the third set at 37° C.

Titrations were made daily with 1-20th N.NaOH to determine the degree of acid developed. The results are expressed in per cent. LACTIC ACID.

Table 7a shows a relation of acid colonies to total numbers. In a general way it can be stated that the higher the total count, the higher the proportion of acid formers. The highest count in sample 4 of 29,810,000 bacteria per cc. all the colonies appeared to be acid colonies. No doubt there were some others, but the acid formation by the large number of acid forming bacteria was so great that other bacteria could not counteract the influence. No doubt, if a much larger number of samples could have been examined the figures would be smoother and clearer. This also applies to table 7. Here all samples were averaged according to the part of the original of ice cream the sample was taken from. In the last column a ratio was computed, which brings all the figures into a simple relation. This ratio indicated that samples B1 to 4 contained the largest numbers of bacteria. These samples were from the surface. Next in number are the samples taken one inch from the surface (C4). Next in line is the number taken three-quarters way down. Why this should be high I have no explanation to offer. Larger numbers of samples would, no doubt, clear up such points. There is also the fact that the most expensive ice cream had small numbers, although some of the cheaper ones were low. In fact sample 6, which cost $1.25, was the lowest.

The counts on dextrose-litmus-agar were always much higher than on plain agar.

Report On Tests of the Relation of Lactic Acid Produced by Streptococcus Lacticus to Numbers of Bacteria and the Relation of Numbers of Bacteria to Taste and Appearance.

Nine flasks (1 liter Erlenmeyer) were sterilized. Into three flasks were placed 400 cc. certified raw milk, into three 400 cc. certified raw cream, and into the last three 400 cc. certified milk were placed and these last three sterilized in the autoclave for fifteen minutes at two atmospheres pressure. All flasks were inoculated with dex-

trose broth cultures of a strain of Streptococcus lacticus. This streptococcus was isolated for this work from ice cream. This fact must be borne in mind as it may have influenced the results to some extent. Immediately after inoculation plates were prepared in plain agar and in dextrose-litmus-agar in dilutions of 1:1000 to determine the number of streptococci inoculated. The incubation temperatures were 37° C., 21° C. and 7° C. One flask with raw milk, one with raw cream and one with sterilized milk placed at each of the three temperatures. Daily tests were made as follows:—

1. The taste and appearance.

2. The acidity was determined by titrating 5 cc. of the inoculated milk or cream, diluted to 50 cc. with distilled water, against one-twentieth normal sodium hydrate solution. The degree of acidity was then calculated in per cent. lactic acid.

3. Plates in duplicates were made in plain agar, and dextrose-litmus-agar the same as with the ice cream excepting that dilutions had to be carried to 1:10.000.000. Six counts were made of these plates after forty-eight hours.

It should be stated here that the colonies formed by this particular strain of Str. lacticus were exceptionally small, so that the agar plates showed a relatively low count and undoubtedly the counts on dextrose-litmus-agar were frequently not as high as they should be. Here also the counts averaged much higher in dextrose-litmus-agar than on plain agar.

Table 7a (See pages 60-61), shows the results in detail.

The figures represent the averages of the duplicate plates, the percentage of acid colonies and the percentage of acid expressed in lactic acid.

At 37° acid formed so rapidly that in twenty-four hours the cream and milk in all cases was curdled. The curd in the sterile milk was smooth and homogeneous, in the

raw milk and cream there was considerable whey. The raw milk at 21° was beginning to show signs of turning after twenty-four hours and contained 185 million bacteria in DLA. The raw cream was still sweet after twenty-four hours, but had curdled after forty-eight hours. It was sweet in spite of the 156 million bacteria after twenty-four hours. The sterile milk was curdled after forty-eight hours. After twenty-four hours it was turning with indications of curd formation (small flakes), and contained 202 million of bacteria. At 7% the souring process and curdling were slow. In the raw milk there was no decided acid taste after nine days, and the bacteria numbered 52 million. A bitter taste had developed. In the raw cream after eight days it seemed to be turning, although the taste was rather bitter than sour. After nine days the taste was decidedly bitter and the bacteria numbered 120 millions. In the sterilized milk the taste was not decidedly sour, and there were no indications of curdling after eight days in spite of 160 millions acid forming bacteria.

To make the results of this more striking I have plotted the results in curves in tables 9, *a*, *b* and *c*, and in 10, *a*, and *c*. Here also I feel that the curves would be smoother and better if a large series of tests were made and with different strains of lactic streptococci.

Table 9*a*—(See page 66.) Raw milk at 37°. The numbers of bacteria are highest after two days, after which there is a falling off. The acidity, however, continues to rise. Several explanations are possible for this. Acid is produced by an enzym, and therefore, will continue even after the bacteria have been removed or decrease. Thirty-seven degrees is a temperature at which such enzyms act more powerfully and more rapidly than at lower temperatures. There is the other explanation which accounts for acid formation in the *apparent* absence of bacteria. Streptococcus lacticus

multiplies only to a certain point. The so-called high acid forming bacilli then produce more lactic acid, *but these bacteria cannot be demonstrated on ordinary media.* It is also possible that this particular strain of Str. lacticus dies sooner than some other strains. In raw milk at 21° the curves are similar to those at 37°, the numbers are higher, but the acidity is lower. High acid formers multiply but slowly at this temperature, so that the acid naturally would be less and enzyms also act slower. At 7° the curves are low and practically parallel. (See page 67.)

Table 9b—The sudden rise in numbers of bacteria in cream at 37° is enormous. The curves at 21° are even, and at 7° low. At 21° the numbers and the amount of acid are nearly parallel. (See page 67.)

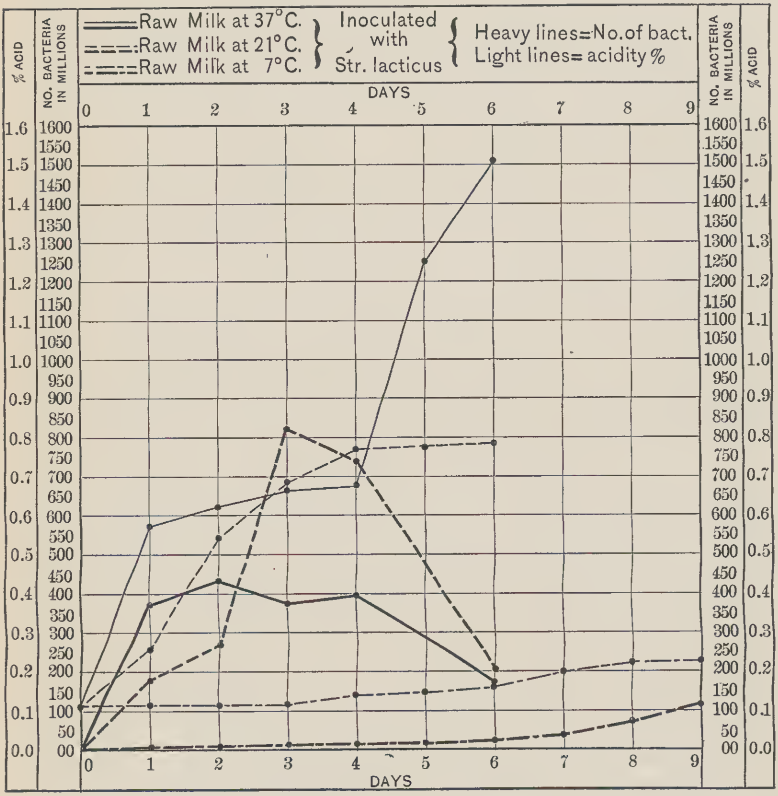
Table 9c—At 7° the respective curves remain parallel. There is no great divergence at either 21° or at 37°. As we are dealing here with pure cultures of the streptococcus, no complicating factors arise through the presence of other bacteria.

Table 10*a*. (See page 69.)—This table shows the relative curves of the three KINDS of flasks at the same temperature. Cream has the greatest number of bacteria, milk next and sterile milk the smallest. The acid formation is highest in milk, next in cream and smallest in sterile milk.

Table 10*b*. (See page 70.)—The acid curves in all three kinds are very consistent. In raw milk there is a decided drop in numbers, in the cream the numbers are relatively small, but are in good accord with the acid formation in the sterile milk.

Table 10*c*. (See page 71.)—The curves are low and smooth.

TABLE 9A



% ACID
NO. BACTERIA IN MILLIONS
Raw Milk at 37°C.
Raw Milk at 21°C.
Raw Milk at 7°C.
Inoculated with Str. lacticus
Heavy lines=No. of bact.
Light lines= acidity %
DAYS
0 1 2 3 4 5 6 7 8 9
1600 1550 1500 1450 1400 1350 1300 1250 1200 1150 1100 1050 1000 950 900 850 800 750 700 650 600 550 500 450 400 350 300 250 200 150 100 50 00
1.6 1.5 1.4 1.3 1.2 1.1 1.0 0.9 0.8 0.7 0.6 0.5 0.4 0.3 0.2 0.1 0.0

TABLE 9B

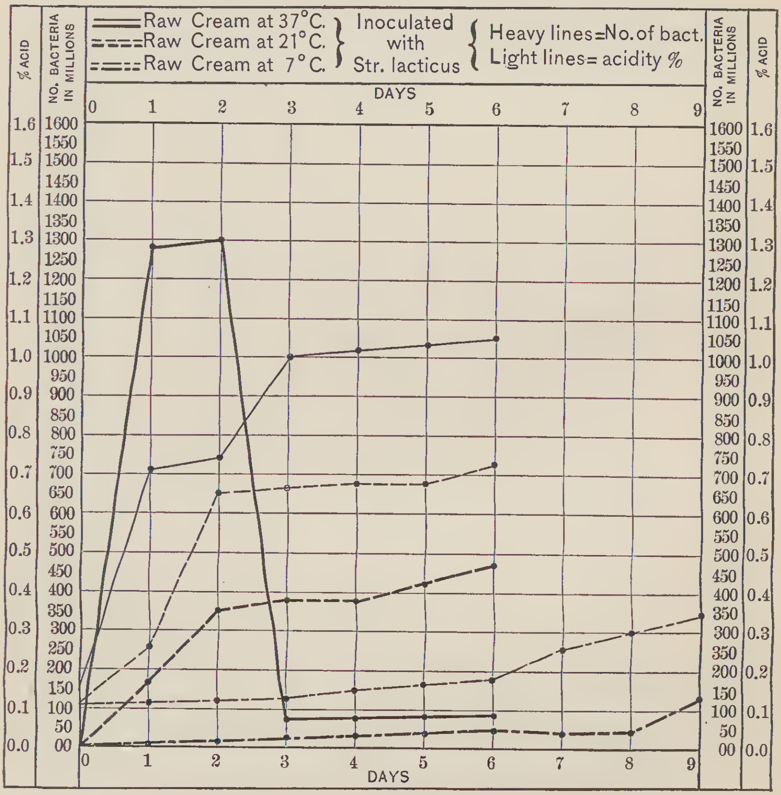
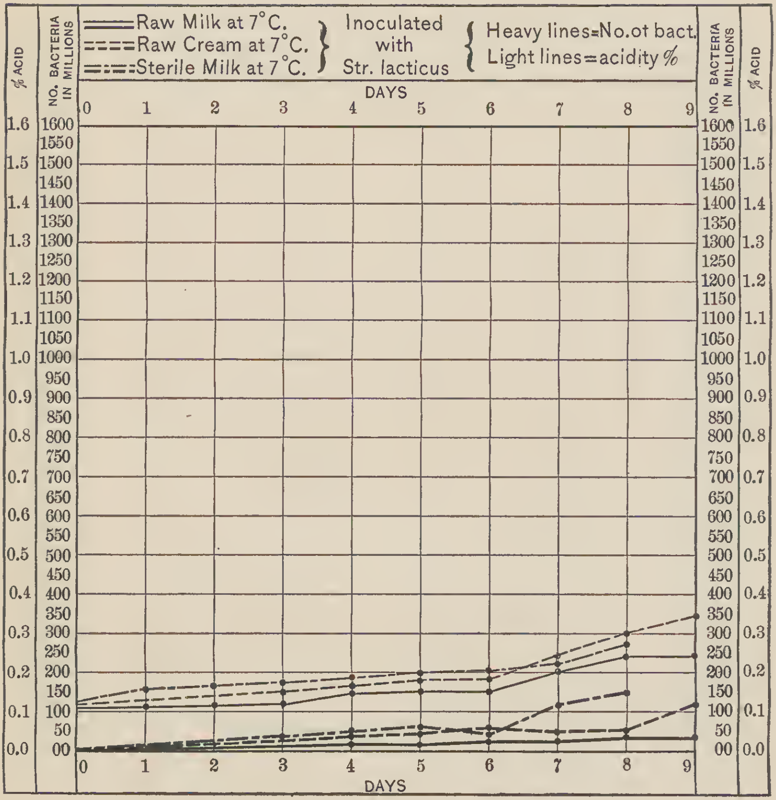
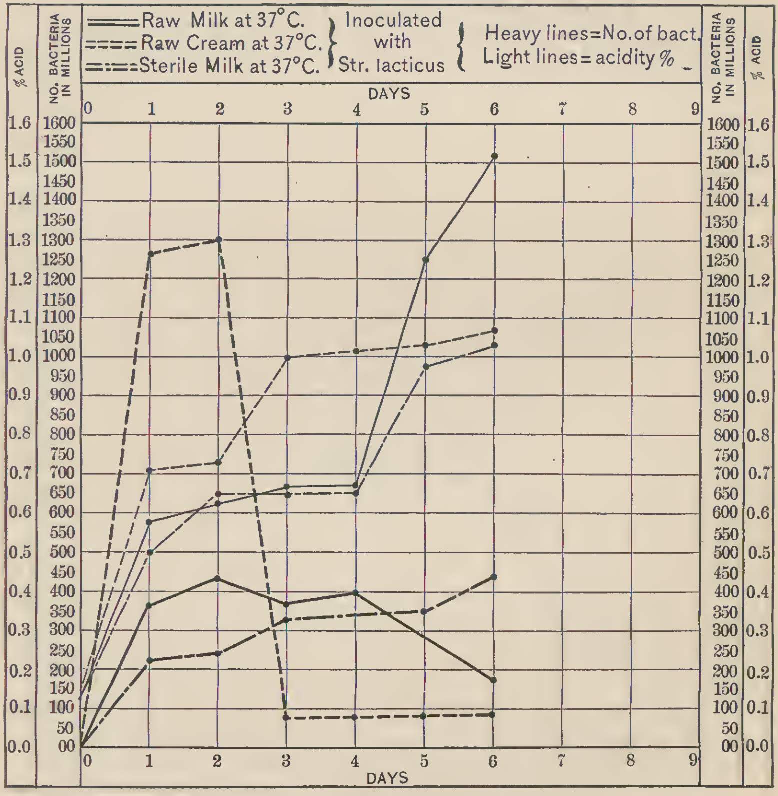

Raw Cream at 37°C.
Raw Cream at 21°C.
Raw Cream at 7°C.
Inoculated with Str. lacticus
Heavy lines=No. of bact.
Light lines= acidity %
% ACID
NO. BACTERIA IN MILLIONS
DAYS
0 1 2 3 4 5 6 7 8 9
1.6 1.5 1.4 1.3 1.2 1.1 1.0 0.9 0.8 0.7 0.6 0.5 0.4 0.3 0.2 0.1 0.0
1600 1550 1500 1450 1400 1350 1300 1250 1200 1150 1100 1050 1000 950 900 850 800 750 700 650 600 550 500 450 400 350 300 250 200 150 100 50 00

TABLE 9C



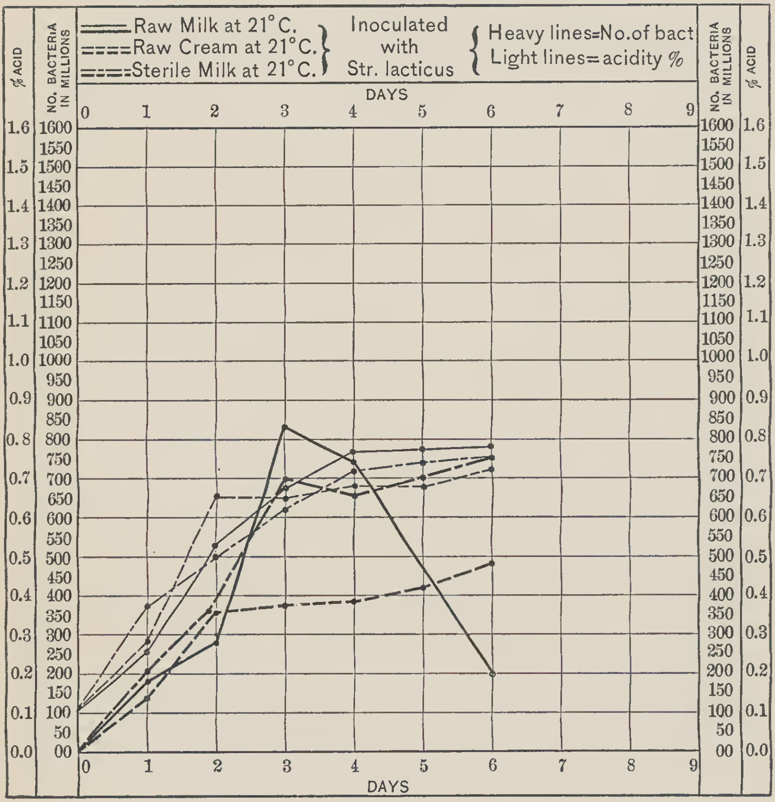
% ACID
NO. BACTERIA IN MILLIONS
Raw Milk at 7°C.
Raw Cream at 7°C.
Sterile Milk at 7°C.
Inoculated with Str. lacticus
Heavy lines=No.of bact.
Light lines=acidity %
DAYS
0 1 2 3 4 5 6 7 8 9
1.6 1.5 1.4 1.3 1.2 1.1 1.0 0.9 0.8 0.7 0.6 0.5 0.4 0.3 0.2 0.1 0.0
1600 1550 1500 1450 1400 1350 1300 1250 1200 1150 1100 1050 1000 950 900 850 800 750 700 650 600 550 500 450 400 350 300 250 200 150 100 50 00

TABLE 10A



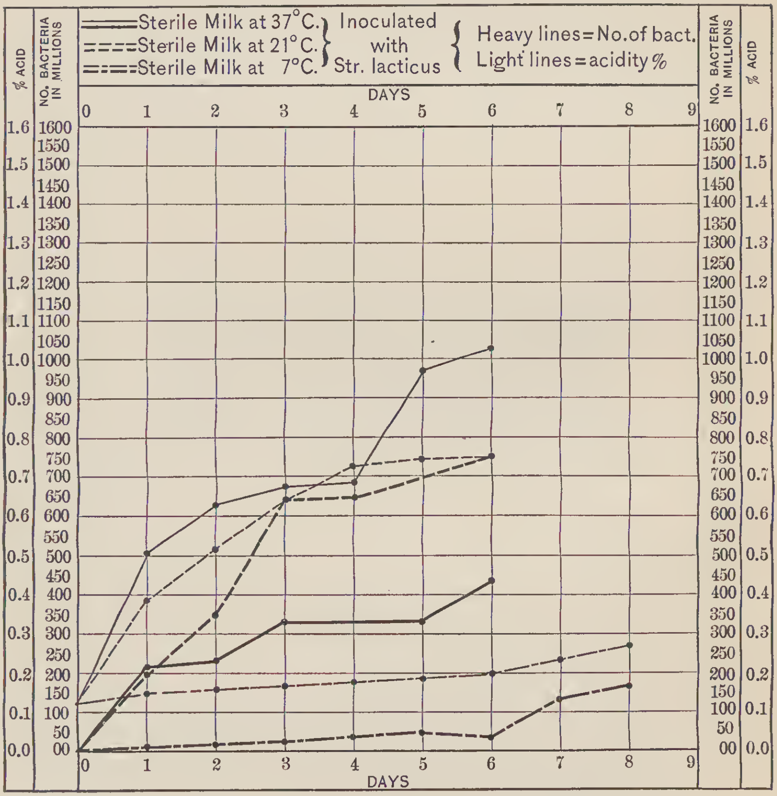
Raw Milk at 37°C.
Raw Cream at 37°C.
Sterile Milk at 37°C.
Inoculated with Str. lacticus
Heavy lines=No. of bact.
Light lines=acidity %
DAYS
% ACID
NO. BACTERIA IN MILLIONS

TABLE 10B



Raw Milk at 21° C.
Raw Cream at 21° C.
Sterile Milk at 21° C.
Inoculated with Str. lacticus
Heavy lines=No. of bact
Light lines=acidity %
% ACID
NO. BACTERIA IN MILLIONS
DAYS

TABLE 10c



Sterile Milk at 37°C.
Sterile Milk at 21°C.
Sterile Milk at 7°C.
Inoculated with Str. lacticus
Heavy lines = No. of bact.
Light lines = acidity %
% ACID
NO. BACTERIA IN MILLIONS
DAYS
0
1
2
3
4
5
6
7
8
9
1600
1550
1500
1450
1400
1350
1300
1250
1200
1150
1100
1050
1000
950
900
850
800
750
700
650
600
550
500
450
400
350
300
250
200
150
100
50
00
1.6
1.5
1.4
1.3
1.2
1.1
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0

Reports Concerning the Significance of Bacterial Counts and Bacillus Coli Tests

H. D. PEASE, M. D.

REPORTS OF BACTERIOLOGICAL INVESTIGATIONS CONCERNING THE SIGNIFICANCE OF BACTERIAL COUNTS AND BACILLUS COLI TESTS OF INDIVIDUAL LOTS OF MARKET ICE CREAM, REFERRED TO AND PRESENTED IN PART AT THE HEARING.

By H. D. Pease, M. D.

Report in the matter of the examination of samples of vanilla and chocolate ice cream purchased from a Commercial Ice Cream Company, New York City, October 22, 1913, and of one sample of Commercial strawberry ice cream, purchased January 31, 1914.

Technique.

Source of Sample. A one gallon can of Vanilla Ice Cream made by a Commercial Ice Cream Company and marked October 22nd, was delivered by them to the Lederle Laboratories.

Method of Taking Samples. Sterile teaspoons were used in digging out the samples and transferring them to sterile 2-oz. and 4-oz. glass stoppered bottles.

Twenty-five to 30 gram samples were taken except in the case of Experiment No. 4. The sample bottles were chilled in ice and salt previous to the taking of the samples. Immediately after taking each sample the bottle was covered with ice and salt so that the ice cream remained frozen until the individual sample was removed from the ice and salt for examination.

Method of Melting Sample Before Examination. A water bath held at 30°C. was used for this purpose.

As soon as the sample was melted it was shaken and allowed to rest for one minute before sampling unless otherwise stated.

Media. 9 liters of Standard Beef Extract Agar were made at one time. Previous to sterilization in the autoclave for 15 minutes at 20 lbs. pressure the agar was

divided among 36 8-oz. flasks. All plates in this series of experiments were poured from this lot of agar.

Dilution Bottles and Water. The bottles used were 4-oz. Philadelphia ovals filled by automatic burettes to contain 99 c.c. of water after sterilization, in the autoclave, 25 minutes at 20 lbs. pressure, and 1-oz. Philadelphia ovals filled in the same manner to contain 9 c.c. of water after the same process of sterilization. The dilution water used contained in all cases 0.5% commercial sodium chloride.

Pipettes. The 1 c.c. pipettes were in all cases straight sided pipettes of even bore and a uniform blunt point. The capacity pipettes were graduated by one mark to contain 1 c.c. of water. The volumetric pipettes were graduated by one mark to deliver 1 c.c. of water.

In reading the amount of liquid in the pipette, care was taken that the end of the pipette rested on the side of the bottle above the sample or dilution in order not to carry over an extra drop of the sample or dilution.

Petri Dishes. 10 x 1½ c.m. sterilized by dry heat at 180°C. for 1½ hours were used for all bacterial counts.

Method of Making Dilutions and Plating. In each case unless otherwise stated the sample of ice cream was measured by a 1 c.c. capacity pipette and the pipette washed out in the dilution water.

The method of dilution was as follows:

1 c.c. of sample	to 99 c.c.	dilution bottle	= 1:100
1 c.c. of 1:100	to 9 c.c.	dilution bottle	= 1:1000
1 c.c. of 1:100	to 99 c.c.	dilution bottle	= 1:10000
1 c.c. of 1:10000	to 9 c.c.	dilution bottle	= 1:100000
1 c.c. of 1:10000	to 99 c.c.	dilution bottle	= 1:1000000

Transfers for higher dilutions were made before 1 c.c. was transferred to the petri dish.

Litmus Lactose Agar Plates. 1 c.c. of a 10% solution of lactose and ½ c.c. of a 1% solution of azolitmin were

added to each plate before inoculation with 1 c.c. of the diluted sample.

Test for Bacteria of the B. coli Type. Durham fermentation tubes were used. The outside tube was 6″ x ⅞″, the inside tube 3″ x ⅜″. The lactose-peptone-bile was made by adding 1% lactose and 1% peptone to fresh ox-bile which had been previously boiled and filtered.

Twelve hundred fermentation tubes were prepared at one time from one lot of bile. Inoculated tubes were incubated 72 hours at 37°C. At the end of each 24-hour period the percentage of gas was recorded. From the two highest dilutions showing gas at the end of 48 hours, litmus-lactose-agar plates were made by transferring one 3 mm. loopful to a 9 c.c. dilution bottle and one 3 mm. loopful from this dilution to a plate containing lactose and azolitmin as described above.

After 24 hours at 37°C. isolations of characteristic colonies were made on lactose agar slants. After 24 hours at 37°C. the growth on the slants were used for the inoculation of tubes containing Durham's peptone solution and nitrate solution. A stab culture was made in the lactose agar slant and in a tube of infusion gelatine. The inoculated peptone solution, nitrate solution and lactose agar stab were incubated 4 days at 37°C.

The peptone solution was tested for the presence of indol by the addition of ½ c.c. of a paradimethylamidobenzaldehyde solution.

The nitrate solution was tested for the presence of nitrites by the addition of ½ c.c. of a sulphanilic acid solution and ½ c.c. of a naphthylamene acetate solution.

The presence of gas in the lactose agar stab was also noted as a control on the isolation.

The gelatine stabs were incubated 10 days at 20°C. and the presence of liquefaction noted.

Bacterial Counts. All plates were counted as far as possible. In reporting the bacterial counts averages were

made in cases where the bacterial counts exceed 20 and less than 500 colonies to the plate. The number reported was not in accordance with the standard method of reporting bacterial milk counts.

Outline of Experiments.

Experiment No. 1. Test of same location (see results reported under A, B, C, D, E and F).

Six samples were taken from as near the same location as possible at a point about 3 inches below the top and at the middle of the can.

Experiment No. 2. Test of separate location.

Six samples reported under G, H, I, J, K, L, were taken as follows:

G—top-side.
H—top-middle.
I—center-side.
J—center-middle.
K—bottom-side.
L—bottom-middle.

Experiment No. 3. Test of triplicate dilutions made from same sample.

The samples taken under Experiment No. 1 were diluted in triplicate and are reported under A_1 A_2 A_3—F_1 F_2 F_3.

Experiment No. 4. Test of the evenness of melting.

Six samples of about 70 c.c. were taken in 4-oz. chemical salt mouth bottles from the side of the can from top to bottom. Each sample was allowed to melt at room temperature (74°F.) for two minutes and the melted portion sampled with a 1 c.c. capacity pipette. These results are reported under M, N, O, P, Q, R.

Experiment No. 5. Test of method of measuring sample for dilution.

Six samples were taken from the same locality at a point immediately surrounding the place where the six samples were taken for Experiment No. 1.

VANILLA ICE CREAM.—EXPERIMENT NO. 1 AND NO. 3 COMBINED.

Six samples (A, B, C, D, E, F) taken from the same location; about three inches from the top and middle of the can.

Multiply results (except B. coli) by 1,000.

	Colony Count 37°—2 days			Litmus-lactose-agar						Gas Production Lac. Pep. Bile							
Sample No.	Dilution	B.E.Agar	Total L.L.Agar	Acid	Inert	Alkal.	% Acid Colony Count	Colony Count 20°-5 da. B.E.Agar	Dilution	24	48	72	Lit. Lac. Agar Plate	Lac. Agar Stab.	Indol	Nitrate	Gelatine
A1	10,000	3,880	1,880	1,620	260		86.3	2,300	100	Tr.	15	20					
	100,000	2,200	800	600	200		75.0	2,200	1,000	Tr.	20	20					
	1,000,000	3,000							10,000	Tr.	20	20	+	=	=	‡	=
									100,000	—	—	—	—				=
A2	10,000	2,100	1,470	1,170	270	30	79.5	2,400	100	Tr.	15	20					
	100,000	2,800	1,000	700	300		70.0	2,900	1,000	Tr.	20	30	+	+	+	+	=
	1,000,000								10,000	Tr.	Tr.	2	—				
									100,000	—	—	—					
A3	10,000	2,870	1,910	1,180	720	10	61.7	2,600	100	Tr.	20	30					
	100,000	2,000	1,800	1,400	300	100	77.0	4,000	1,000	Tr.	15	25	+	=?	=	‡	=
	1,000,000	2,000							10,000	—	—	—					
									100,000	—	Tr.	20	—				
B1	10,000	2,540	1,600	1,550	130	20	96.8	2,300	100	2	40	50					
	100,000	6,800	1,600	1,400	200		87.5	2,700	1,000	—	10	10					
	1,000,000	4,000							10,000	—	20	20	+	=	=	‡	=
									100,000	—	Tr.	2					
B2	10,000	2,450	1,350	1,180	170		87.4	2,600	100	5	30	30					
	100,000	2,600	1,700	1,300	400		76.4	3,000	1,000	Tr.	30	25					
	1,000,000	3,000							10,000	—	20	25	+	—	—	+	—
									100,000	—	Tr.	4	—				—
B3	10,000	spreader	1,100	990	90	20	90.0	2,700	100	Tr.	15	15					
	100,000	1,400	600	500	100		86.5	2,800	1,000	Tr.	15	15	+	—	—	+	—
	1,000,000	4,000							10,000	—	15	15	+	—	—	+	—
									100,000	—	—	—					
C1	10,000	2,360	1,110	740	340	30	66.6	2,700	100	Tr.	30	20	+	+	—	+	—
	100,000	2,800	1,200	800	400		66.6	4,400	1,000	Tr.	20	20	+	—	—	+	—
	1,000,000	6,000							10,000	—	—	—					
									100,000	—	Tr.	2					
C2	10,000	2,380	740	590	140	10	79.7	3,200	100	Tr.	15	20					
	100,000	2,400	800	600	200		66.6	4,000	1,000	Tr.	15	20	+	—	—	+	=
	1,000,000	1,000							10,000	—	15	20	—				
									100,000	—	—	—					
C3	10,000	2,430	810	720	80	10	88.8	2,700	100	Tr.	20	20					
	100,000	2,100	800	500	300		63.3	2,200	1,000	Tr.	10	10	+	—	—	+	—
	1,000,000	5,000							10,000	Tr.	Tr.	2	+	=	=	‡	=
									100,000	—	—	—					

D1	10,000	3,240	1,080	1,020	50	10	94.4	3,000	100	Tr.	20	20					
	100,000	1,700	1,800	1,400	400		87.5	2,600	1,000	Tr.	25	30	+	+	=	—	=
														—		+	
	1,000,000	2,000							10,000	—	30	40	+	+	—	+	=
									100,000	—	—	—					
D2	10,000	1,480	610	510	90	10	83.6	3,000	100	Tr.	20	20	+	—	—	+	—
	100,000	3,200	800	600	200		66.6	3,300	1,000	Tr.	50	40	+	+	+	+	=
	1,000,000								10,000	—	20	25					
									100,000	—	10	30					
D3	10,000	1,700	770	690	60	20	89.6	2,600	100	Tr.	50	50					
	100,000	1,800	1,300	1,300			100.0	2,900	1,000	Tr.	20	30	+	—	—	+	—
	1,000,000								10,000	—	10	10	+	=	=	‡	=
																	—
									100,000	—	—	—					
E1	10,000	2,080	540	460	70	10	85.1	3,000	100	Tr.	20	20	+	—	—	+	—
	100,000	1,800	1,100	800	300		72.8	2,900	1,000	Tr.	20	30	+	=	=	‡	=
	1,000,000	2,000							10,000	—	—	—					
									100,000	—	—	Tr.					
E2	10,000	1,980	790	720	70		91.1	2,600	100	Tr.	20	20					
	100,000	1,600	1,000	700	300		70.0	3,000	1,000	Tr.	40	50	+	+	—	+	—
	1,000,000	3,000							10,000	Tr.	30	40	+	=	=	‡	=
																	—
									100,000	—	—	—					
E3	10,000	1,670	750	620	130		82.6	2,800	100	Tr.	30	30					—
	100,000	1,600	800	600	200		75.0	3,400	1,000	Tr.	20	20	+	—	—	+	—
	1,000,009	1,000							10,000	—	30	50	+	=	=	‡	=
									100,000	—							
F1	10,000	2,070	750	700	50		93.3	2,700	100	Tr.	20	30					
	100,000	1,900	500	400	100		80.0	3,000	1,000	Tr.	20	20					
	1,000,000								10,000	—	20	20	+	—	—	+	—
									100,000	—	Tr.	15	+	+	—	+	—
F2	10,000	1,380	510	410	90	10	80.4	2,700	100	Tr.	30	30					
	100,000	2,200	500	400	100		80.0	2,900	1,000	Tr.	30	20	+	—	—	+	—
	1,000,000	4,000							10,000	Tr.	50	60	+	—	—	+	
									100,000	—	—	—					
F3	10,000	1,300	680	590	80	10	86.7	2,200	100	Tr.	20	25					
	100,000	900	400	200	200		50.0	3,000	1,000	Tr.	20	25					
	1,000,000	2,000							10,000	Tr.	50	75	+	{ =	=	‡	=
														+	—	+	—
									100,000	—	—	—					

Each of the six samples was examined in the following way:

1. One gram of frozen cream weighed and 99 c.c. of water added.

2. One c.c. of liquid not containing air bubbles removed with 1 c.c. capacity pipette.

3. One c.c. of liquid not containing air bubbles removed with 1 c.c. volumetric pipette.

4. One c.c. of liquid containing air bubbles removed with 1 c.c. capacity pipette.

5. One c.c. of liquid containing air bubbles removed with 1 c.c. volumetric pipette.

These results are reported under S_1 S_2 S_3 S_4 S_5—X_1 X_2 X_3 X_4 X_5.

VANILLA ICE CREAM.—EXPERIMENT NO. 2.

Multiply results (except B. coli) by 1,000.

Description of Sample	Sample No.	Dilution	Colony Count 37°—2 days B.E.Agar	Total L.L.Agar	Litmus-lactose-agar Acid	Inert	Alkal.	% Acid Colony Count	Colony Count 20°-5 da. B.E.Agar	Dilution	Gas Production Lac. Pep. Bile 24	48	72	Lit. Lac. Agar Plate	Lac. Agar Stab.	Indol	Nitrate	Gelatine
Top Side	G	10,000	1,700	650	580	70		89.2	2,600	100	Tr.	10	10					
		100,000	1,300	1,100	800	300		72.7	2,800	1,000	Tr.	10	10	+	—	—	+	=
		1,000,000	2,000							10,000	—	Tr.	2	+	—	—	+	—
										100,000	—	—	—					
Top Middle	H	10,000	1,690	700	640	40	20	91.4	2,700	100	Tr.	20	20					
		100,000	1,100	400	300	100		75.	3,400	1,000	—	10	10					
										10,000	—	40	50	+	—	=	‡	=
		1,000,000	2,000												+			
										100,000	—	Tr.	25	+	No growth			
Centre Side	I	10,000	1,910		Alkaline spreader				2,400	100	Tr.	30	40	+	—	+	+	—
		100,000	1,900	600	500	100		83.3	2,500	1,000	Tr.	20	20	+	—	—	+	—
		1,000,000								10,000	—	—	—					
										100,000	—	—	—					
Centre Middle	J	10,000	2,000	800	680	120		85.	2,500	100	Tr.	30	50					
		100,000	1,100	1,200	1,000	200		83.3	2,400	1,000	Tr.	40	50	+	+	+	+	=
		1,000,000	4,000							10,000	—	20	25	+	—	—	+	—
										100,000	—	—	—					
Bottom Side	K	10,000	1,530	880	770	100	10	85.5	2,600	100	Tr.	30	40					
		100,000	700	600	500	100		83.3	2,300	1,000	Tr.	15	15	+	+	?	+	—
		1,000,000	6,000							10,000	—	10	10	+	—	—	+	—
										100,000	—	—	—					
Bottom Middle	L	10,000	3,280	2,240	1,890	340	10	84.3	2,600	100	Tr.	20	20					
		100,000	2,600	1,800	1,400	400		77.7	2,300	1,000	Tr.	30	30	—				
		1,000,000	14 con-							10,000	—	—	—					
			tam.?							100,000	—	10	15	+	+	—	+	—

VANILLA ICE CREAM.—EXPERIMENT NO. 4.

Six samples of about 70cc taken from the side of the can from top to bottom.

Multiply results (except B. coli) by 1,000.

Sample No.	Dilution	Colony Count 37°—2 days: B.E.Agar	Total L.L.Agar	Litmus-lactose-agar: Acid	Inert	Alkal.	% Acid Colony Count	Colony Count 20°-5 da. B.E.Agar	Dilution	Gas Production Lac. Pep. Bile: 24	48	72	Lit. Lac. Agar Plate	Lac. Agar Stab.	Indol	Nitrate	Gelatine
M	10,000	2,620	2,370	1,920	440	10	81.0	2,320	100	Tr.	15	15	+	$\underline{+}$	=	‡	=
	100,000	2,400	2,200	2,000	200		90.9	1,900	1,000	Tr.	10	10	—				
	1,000,000	4,000							10,000	—	—	—					
									100,000	—	—	—					
N	10,000	1,890	1,560	1,290	260	10	82.7	1,700	100	Tr.	20	20					
	100,000	1,400	2,000	1,600	400		80.	1,000	1,000	Tr.	5	10	+	—	—	+	—
	1,000,000	3,000							10,000	—	20	30	+	—	—	+	=
									100,000	—	—						
O	10,000	4,070	1,200 est. alkaline spreader					2,680	100	Tr.	20	15					
	100,000	3,100	1,800	1,400	400		77.7	2,900	1,000	Tr.	20	15	+	‡	=	‡	—
	1,000,000	3,000							10,000	—	10	15	—				
									100,000	—	—						
P	10,000	2,360	1,470	1,360	100	10	92.5	2,600	100	5	30	30					
	100,000	900	1,400	1,300	100		92.8	2,400	1,000	5	30	40	+	‡	=	‡	$\underline{=}$
	1,000,000	1,000							10,000	—	10	10	+	‡	=	‡	=
									100,000	—	—	—					
Q	10,000	2,920	2,010	1,720	270	20	85.5	2,200	100	Tr.	20	25					
	100,000	3,100	500	500			100.	1,200	1,000	Tr.	20	20	+	+	—	+	—
	1,000,000	2,000							10,000	—	20	25	—	—	—	+	
									100,000	—	10	30	+	—	—	+	=
R	10,000	2,050	1,940	1,560	380		80.4	2,000	100	Tr.	30	30					
	100,000	1,500	13,200	12,000	1,200		90.9	1,100	1,000	Tr.	30	30	+	=	=	‡	=
	1,000,000	2,000							10,000	Tr.	40	50	+	—	—	+	—
									100,000	—	—	—					

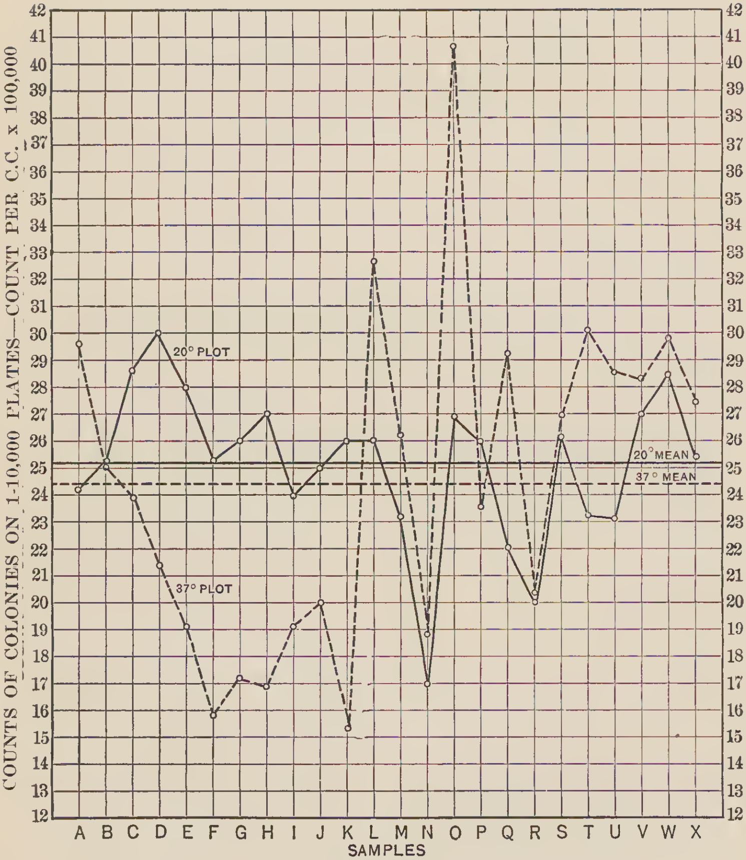


CHART NO. 1.

Plot of the 20°C. and 37°C. beef extract agar bacterial counts of Vanilla Ice Cream samples, made from the 1-10,000 dilutions; and of the means of these counts.

VANILLA ICE CREAM.—EXPERIMENT NO. 5.

Six samples (S, T, U, V, W, X) taken from the same locality at a point adjacent to the place where the six samples for Exp. No. 1 were taken, 3 inches from top near the middle of the can.

Multiply results (except B. coli) by 1,000.

Sample No.	Dilution	Colony Count 37°—2 days B.E.Agar	Total L.L.Agar	Litmus-lactose-agar Acid	Inert	Alkal.	% Acid Colony Count	Colony Count 20°-5 da. B.E.Agar	Dilution	Gas Production Lac. Pep. Bile 24	48	72	Lit. Lac. Agar Plate	Lac. Agar Stab.	Indol	Nitrate	Gelatine
S1	10,000	2,770	1,610 ?	880 ?	730 ?	alk.spdr.	54.6	2,300	100	Tr.	10	10					
	100,000	2,100	2,300	1,800	400	100	78.1	2,900	1,000	Tr.	10	10	+	=	=	‡	=
	1,000,000	2,000							10,000	—	10	10	+	+	+	+	—
									100,000	—	—	—					
S2	10,000	3,460	2,640	2,220	400	20	84.0	2,600	100	Tr.	10	10					
	100,000	2,100	1,300	900	400		69.2	3,500	1,000	—	5	5					
	1,000,000	8,000							10,000	—	2	2	+	—	—	+	—
									100,000	—	Tr.	25	+	±	=	‡	=
S3	10,000	2,130 spdr.	1,460	760	700		52.0	3,100	100	Tr.	20	20	+	—	—	+	=
	100,000	2,700	1,800	1,200	500	100	66.6	2,700	1,000	—	10	10	+	—	—	+	=
	1,000,000	3,000							10,000	—	—	—					
									100,000	—	—	—					
S4	10,000	2,920	2,250	1,700	540	10	75.5	3,000	100	2	40	50					
	100,000	2,600	1,400	1,000	400		71.4	2,400	1,000	—	10	10	+	—	—	+	—
	1,000,000								10,000	—	Tr.	2	—				
									100,000	—	—	—					
S5	10,000	2,280	2,000 est. alkaline spreader					2,100	100	10	30	40					
	100,000	2,200	2,400	1,600	800		66.6	2,400	1,000	Tr.	20	20	+	—	—	+	—
	1,000,000	4,000							10,000	—	Tr.	2	—				
									100,000								
T1	10,000	3,000	2,600	2,000	530	70	76.9	2,500	100	10	30	40					
	100,000	2,700	1,600	1,200	400		75.0	2,700	1,000	Tr.	40	50	+	+	—	+	—
	1,000,000	6,000							10,000	—	Tr.	2	+	—	—	+	—
									100,000	—	—	—					
T2	10,000	2,570	2,770	1,950	780	40	70.3	2,200	100	2	30	40					
	100,000	3,300	1,600	1,200	400		75.0	3,500	1,000	Tr.	20	30					
	1,000,000	5,000							10,000	—	20	30					
									100,000	—	10	10	+	—	—	+	—
T3	10,000	2,480	2,180	1,560	600	20	87.9	2,100	100	Tr.	10	20					
	100,000	3,300	2,500	1,900	500	100	76.0	4,000	1,000	Tr.	10	20	+	—	—	+	—
	1,000,000	10,000							10,000	—	10	15	—				
									100,000	—	—	—					
T4	10,000	3,460	3,200 est.	2,800 est.	400 est.	alk.spdr. over ½ plate	87.5 ?	2,200	100	5	20	30	+	+	+	+	—
	100,000	3,600	2,800	2,200	600		78.5	3,100	1,000	Tr.	10	15	+	—	—	+	+
	1,000,000	6,000							10,000	—	—	Tr.					
									100,000	—	—	—					
T5	10,000	3,590	1,200 est.	1,200 est.	alk spdr.		100. ?	2,600	100	30	50	60					
	100,000	3,500	2,400	1,800	600		75.0	3,000	1,000	Tr.	30	60	+	+	+	+	—
	1,000,000	10,000							10,000	—	40	60	+	—	—	+	—
									100,000	—	—	—					

U1	10,000	2,910	2,170	1,620	530	20	74.6	2,400	100	2	30	30				
	100,000	2,300	2,900	2,400	500		82.7	3,500	1,000	2	80	90	+	+	—	+	—
	1,000,000	5,000							10,000	—	30	30	+	—	—	+	—
									100,000	—	—	—					
U2	10,000	2,780		alk.	spreader			2,750	100	5	30	30					
	100,000	2,700	1,500	800	700		53.3	3,500	1,000	Tr.	10	10					
	1,000,000	1,000							10,000	Tr.	25	30	+	—	—	+	—
									100,000	—	Tr.	10	—				
U3	10,000	2,430	2,000 est.	2,000 est.	alk. spdr.		100.?	1,600	100	Tr.	30	50					
	100,000	3,400	1,600	1,000	600		47.0	2,400	1,000	Tr.	20	30					
	1,000,000	6,000							10,000	—	20	30	+	—	—	+	—
									100,000	—	Tr.	Tr.	—				
U4	10,000	3,580	2,800 est.	2,200 est.	600 est.alk.spd.		71.4?	2,800	100	Tr.	10	15					
	100,000	2,800	2,400	2,200	200		91.6	3,800	1,000	Tr.	10	15	+	+	—	+	—
	1,000,000	2,000							10,000	—	Tr.	2	+	—	—	+	—
									100,000	—	—	—					
U5	10,000	2,620	2,470	1,910	550	10	77.3	2,100	100	10	30	40					
	100,000	2,100	2,400	1,800	600		75.0	2,500	1,000	Tr.	10	15	+	—	—	+	—
	1,000,000	1,000							10,000	—	5	10	+	=	=	‡	—
									100,000	—	—	—					
V1	10,000	2,380	1,640	1,060	580		64.6	2,000	100	2	40	50					
	100,000	2,800	1,500	900	600		60.0	2,400	1,000	Tr.	30	40	+	=	‡	‡	=
	1,000,000	2,000							10,000	Tr.	40	50	+	—	—	+	—
									100,000	—	—	—					
V2	10,000	3,440	1,990	1,390	560	40	69.7	3,600	100	Tr.	20	20					
	100,000	4,800	2,600	1,600	1,000		61.5	5,000	1,000	Tr.	10	10					
	1,000,000	8,000							10,000	—	30	30	—				
									100,000	—	5	15					
V3	10,000	3,050	3,030	1,560	460	10	51.4	3,500	100	Tr.	50	60					
	100,000	3,500	1,300	1,000	300		76.9	3,000	1,000	Tr.	20	20	+	—	—	+	—
	1,000,000	2,000							10,000	—	10	10	+	—	—	+	—
									100,000	—	—	—					
V4	10,000	2,880	1,770	1,270	480	20	71.7	2,400	100	Tr.	30	30					
	100,000	2,400	1,600	1,200	400		75.0	3,000	1,000	Tr.	40	50					
	1,000,000	1,000							10,000	Tr.	20	20	—				
									100,000	—	10	20	—				
V5	10,000	2,380	1,790	1,130	660		63.1	2,200	100	2	30	30					
	100,000	2,000	1,000	600	400		60.0	2,000	1,000	—	10	10	+	—	—	+	—
	1,000,000	2,000							10,000	—	10	15	+	—	—	+	—
									100,000	—	—	—					

VANILLA ICE CREAM.—EXPERIMENT NO. 5.—(Continued)

Sample No.	Dilution	Colony Count 37°—2 days: B.E.Agar	Total L.L.Agar	Litmus-lactose-agar: Acid	Inert	Alkal.	% Acid Colony Count	Colony Count 20°-5 da. B.E.Agar	Dilution	Gas Production Lac. Pep. Bile: 24	48	72	Lit. Lac. Agar Plate	Lac. Agar Stab.	Indol	Nitrate	Gelatine
W1	10,000	2,670	1,710	1,170	540		68.4	2,100	100	Tr.	20	20					
	100,000		100		100			400	1,000	Tr.	40	50	+	—	—	+	—
	1,000,000	1,000							10,000	—	30	30					
									100,000	—	—	—					
W2	10,000	3,180	2,430	1,720	650	60	70.7	2,750	100	5	20	30					
	100,000	2,800	1,500	1,000	500		66.6	3,000	1,000	Tr.	20	20	+	—	—	+	—
	1,000,000								10,000	—	—	—					
									100,000	—	—	—					
W3	10,000	2,660	1,950	1,420	520	10	72.8	3,000	100	Tr.	30	30					
	100,000	3,100	1,900	1,400	500		73.6	2,000	1,000	50	50	50	+	+	—	+	—
	1,000,000	4,000							10,000	—	—	—					
									100,000	—	10	20	—				
W4	10,000	2,690	2,360	1,720	620	20	72.8	3,200	100	Tr.	15	20					
	100,000	2,500	3,500	no litmus in plate				2,800	1,000	Tr.	15	20	+	=	=	‡	—
	1,000,000	2,000							10,000	Tr.	40	40	+	—	—	+	—
									100,000	—	—	—					
W5	10,000	3,730	3,180	2,190	950	40	68.8	3,200	100	5	20	30					
	100,000	3,000	3,500	2,700	700	100	77.1	4,000	1,000	Tr.	10	15					
	1,000,000	7,000							10,000	—	30	30	+	—	—	+	—
									100,000	—	5	10	—				
X1	10,000	2,910	2,300	1,550	740	10	67.3	2,600	100	5	40	40					
	100,000	3,200	1,700	1,200	400	100	70.5	2,000	1,000	Tr.	20	20	+	=	=	‡	—
	1,000,000	3,000							10,000	Tr.	60	60					
									100,000	—	—	—					
X2	10,000	2,740	2,320	1,760	510	10	75.8	2,600	100	5	20	20					
	100,000	2,200	4,300	3,800	500		88.3	2,600	1,000	Tr.	10	10	+	—	—	+	—
	1,000,000	2,000							10,000	Tr.	15	20	+	—	—	+	—
									100,000	—	—	—					
X3	10,000	2,820	2,730	2,090	590	500	76.5	2,600	100	Tr.	50	50					
	100,000	3,000	2,900	2,000	900		62.0	2,300	1,000	Tr.	20	30	+	—	—	+	—
	1,000,000	1,000							10,000	Tr.	70	80	+	+	—	+	—
									100,000	—	—	—					
X4	10,000	2,230	3,050	2,040	1,000	10	66.8	2,400	100	Tr.	10	10					
	100,000	2,500	3,100	2,000	1,100		64.5	3,000	1,000	Tr.	20	20	+	—	—	—	—
	1,000,000	2,000							10,000	—	10	15	+	=	=	‡	=
									100,000	—	—	Tr.					
X5	10,000	3,000	2,690	2,070	600	20	76.8	2,500	100	Tr.	20	30					
	100,000	2,400	1,600	1,200	400		75.0	2,800	1,000	—	20	30	+	+	—	+	—
	1,000,000	3,000							10,000	—	10	15	+	—	—	+	—
									100,000	—	—	—					

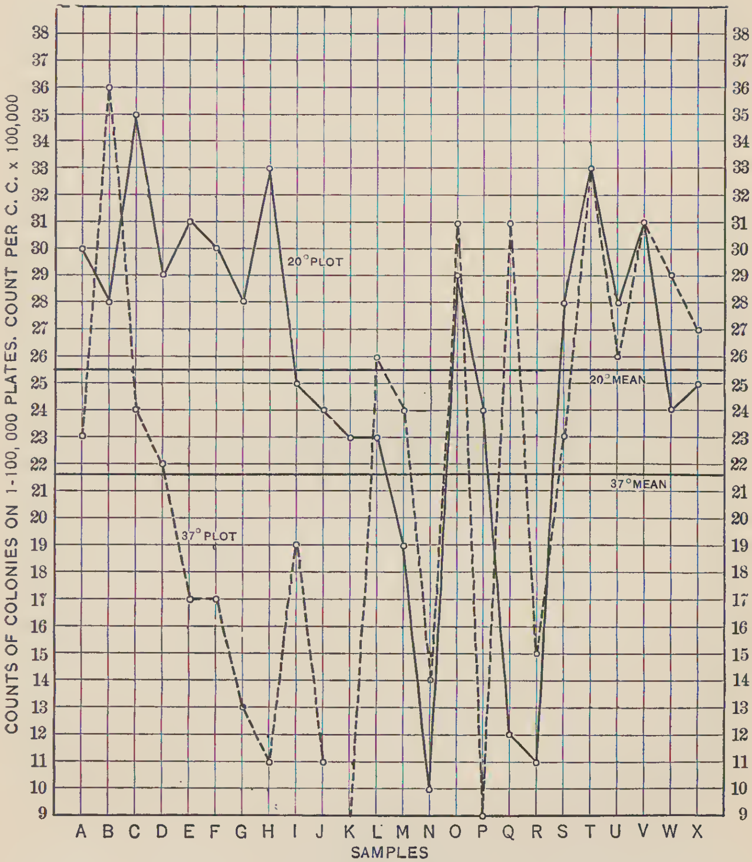


CHART NO. 2.

Plot of the 20°C. and 37°C. beef extract agar bacterial counts of Vanilla Ice Cream samples, made from the 1-100,000 dilutions; and of the means of these counts.

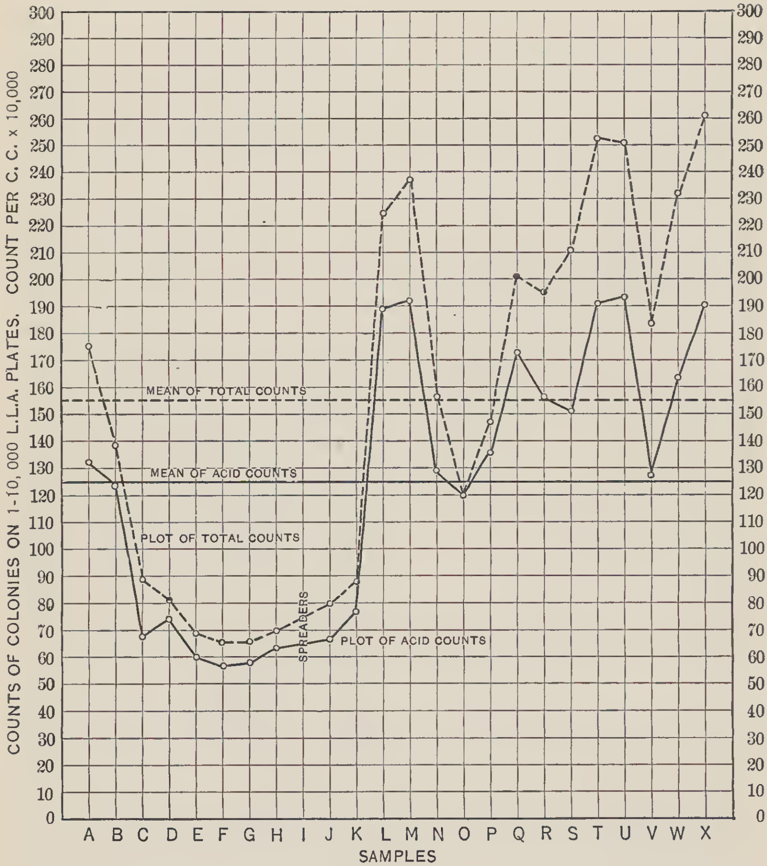


CHART NO. 3.

Plot of the 37°C. litmus lactose agar total and acid forming bacterial counts of Vanilla Ice Cream samples, made from the 1-10,000 dilutions; and of the means of these counts.

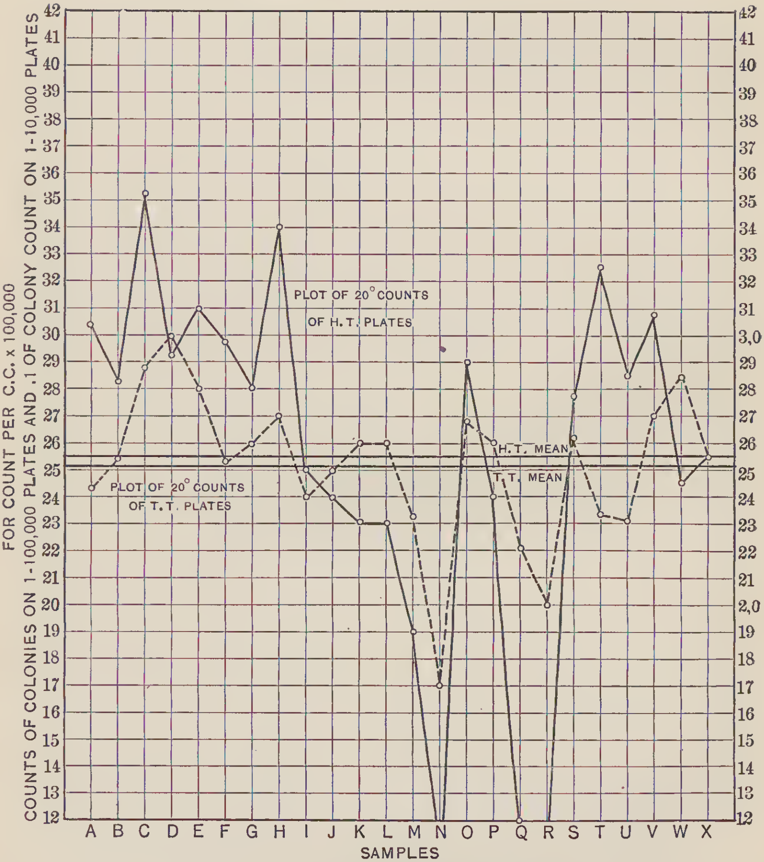


CHART NO. 4.

Plot of the 20°C. beef extract agar bacterial counts of Vanilla Ice Cream samples, made from the 1-10,000 and 1-100,000 dilutions of same samples; and of the means of these counts.

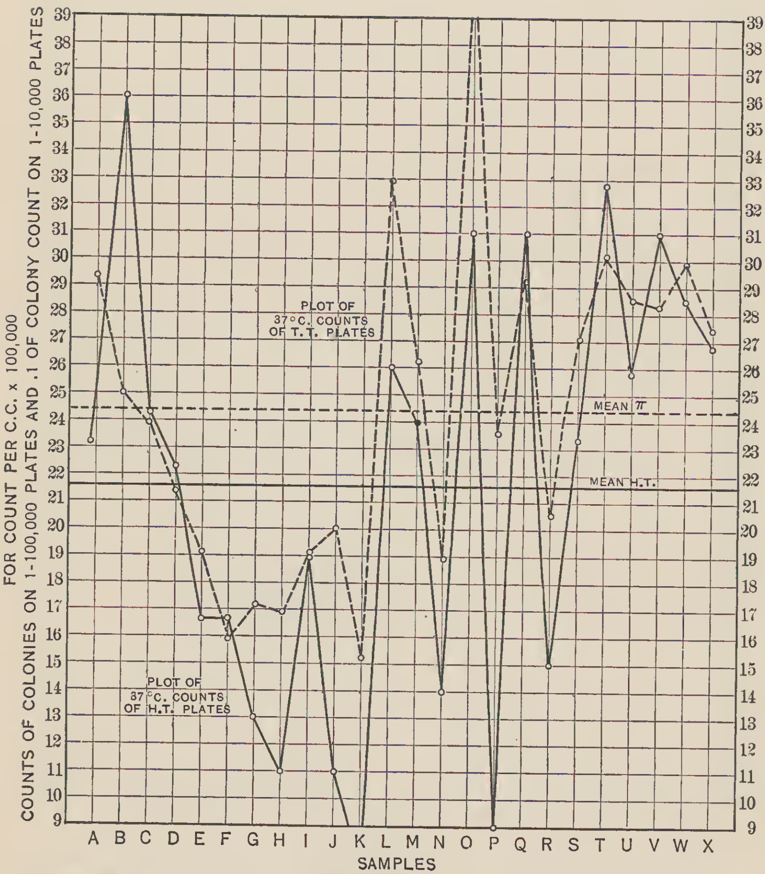


CHART NO. 5.

Plot of the 37°C. beef extract agar bacterial counts of Vanilla Ice Cream samples, made from 1-10,000 and 1-100,000 dilutions of the same samples; and of the means of these counts.

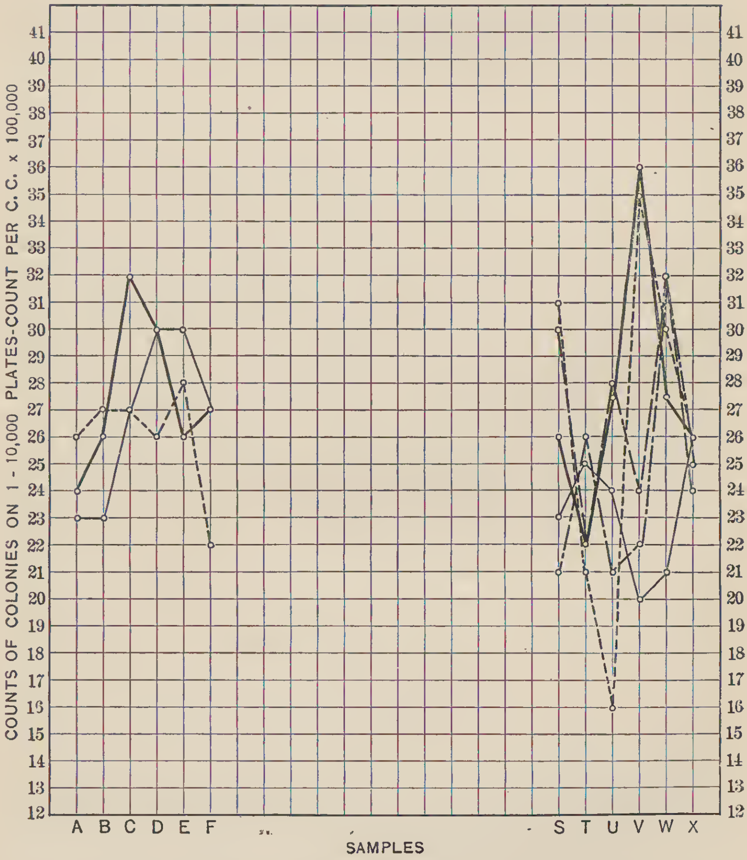


CHART NO. 6.

Plot of the 20°C. beef extract agar counts of Vanilla Ice Cream samples made from the 1-10,000 dilutions.

Samples A to F—Tests made in triplicate by the same method.

Samples S to X, inclusive—Tests made by different methods of sampling.

Key to Plot of Samples.

Heavy solid line—Plot of Sample examined by Method No. 1.

Light solid line—Plot of Sample examined by Method No. 2.

Dotted line—Plot of Sample examined by Method No. 3.

Dash line—Plot of Sample examined by Method No. 4.

Dash and dot line—Plot of Sample examined by Method No. 5.

Method No. 1. One gram of frozen cream weighed out and 99 c.c. water added.

Method No. 2. One c.c. melted ice cream not containing air bubbles removed with 1 c.c. capacity pipette.

Method No. 3. One c.c. melted ice cream not containing air bubbles removed with 1 c.c. volumetric pipette.

Method No. 4. One c.c. melted ice cream containing air bubbles removed with 1 c.c. capacity pipette.

Method No. 5. One c.c. melted ice cream containing air bubbles, removed with 1 c.c. volumetric pipette.

Samples A, B, C, D, E, F, S, T, U, V, W, X all removed from same immediate location.

Vanilla Ice Cream.

Comments on Results of Tests as Shown by Charts 1 to 6 (Inclusive).

CHART NO. 1.

1. The results of the tests on samples A to F (inclusive), obtained from one portion of the can, as given for the 1 to 10,000 dilution at either of the two temperatures, show marked numerical differences, notwithstanding that these results are the averages of triplicate tests on each sample. The results of any individual test are not harmonious with the results of any of the others, as is shown for the 20°C. tests under A to F (inclusive) in Chart No. 6.

2. The results on these six samples at 37°C. showed

much greater variations than did those obtained from the 20°C. tests.

3. The variations in the results on these six samples from the one location, at 20°C., as already noted, were greater than those in the results of the tests of the following six samples, G to L (inclusive), obtained from various portions of the can contents, when tested at the same temperature; but the reverse is true of the results of the tests at 37°C.

4. In the tests of samples M to R (inclusive), made for the purpose of determining evenness of melting, there occurred the most extreme differences to be noted in any of the comparisons of results on this chart. While a general parallelism between the 20°C. and 37°C. counts was to be noted, the results of the tests made at the latter temperature were almost invariably higher than the former. This reverses the results of the tests on samples A to L, all of which were melted in the same manner.

5. A greater degree of uniformity is to be noted in the results obtained in the case of samples S to X (inclusive), which samples were tested by five different methods of examination. This uniformity, however, is due, in large part, to the fact that these results are averages of those obtained by the application of the five different methods of examination. The full details of these differences are shown under samples S to X in Table 6.

There is noted a parallelism between the 20°C. and the 37°C. counts on these samples, S to X; but the counts at the latter temperature were higher, which is a reversal of the results obtained from the tests of samples A to F, which samples were taken from the same portion of the can.

CHART NO. 2.

6. The results of the counts of the 1 to 100,000 dilution plates on all the samples showed generally greater variations than those obtained from the 1 to 10,000 dilution

plates shown in Chart 1. This is clearly brought out by noting the greater difference between the means of the counts at 20°C. and those at 37°C. in Chart 2, as compared with the means in Chart 1.

7. There is a general resemblance between the outlines of the plottings of Chart 2 when compared with those on Chart 1, but upon studying the results in each chart for any one of several particular samples great variations are to be found. This is well illustrated by a study of sample P.

8. Upon attempting to compare the details of Chart 2, as has been done in comments 2, 3, 4, and 5 for Chart 1, we find that the same statements would be inappropriate for this chart. In some instances the comments would be accurately descriptive of the results, but in other instances the reverse is true.

CHART NO. 3.

9. A comparison of the results of the counts on separate samples, when tested upon litmus lactose agar, using the plates of the 1 to 10,000 dilution, with the counts of acid colonies on the same plates shows a marked parallelism between the two.

10. The plottings in this chart show a general parallelism with the 37°C. plottings in Charts 1 and 2, except for the samples K to X. Among these samples we find some of the plottings similar to those in the previous charts, but others are quite unlike them.

CHART NO. 4.

11. In this chart, comparing the results of counts of dilution plates 1 to 10,000 with those at 1 to 100,000, incubated at the one temperature (20°C.), we find an apparent parallelism between the plottings; but upon close examination we find sometimes the 1 to 10,000 plates giving

the higher results, and at other times the 1 to 100,000 plates showing the higher counts.

12. The average results of the triplicate samples, as shown for A to F (inclusive), together with the results of tests of samples from scattered locations in the can, C to L (inclusive), show a greater degree of uniformity than do the results of the evenness of melting samples M to R. The results in the latter group of samples greatly differ from those in the former in two general locations and are also quite at variance with the results of the averages of the samples S to X.

13. The averages of the results of the application of the five different methods to samples S to X (inclusive), showing variations of 1,000,000 in a maximum bacterial count of but little over 3,000,000, show clearly the unreliability even of averages of five samples when the methods are somewhat at variance. These variations are, however, but little greater than the variations to be found in the averages of three tests in which the same method was employed, as is shown by the plottings of the results of samples A to F in this table. It cannot be said, therefore, that the differences in methods were the causes of the variations in the results obtained in samples S to X. Apparently slight variations in method do not lead to differences in bacterial counts of such magnitude as are liable to be caused by the natural variations in the bacterial content of the ice cream itself.

CHART NO. 5.

14. In this chart we have the most extreme variations noted in any comparison of results from the examination of the samples of the vanilla product. This greater variation is an expression of the result that has been obtained in all of the examinations of ice cream, namely, that in comparing the 37° counts with similar counts at

20°, which are plotted in chart 4 in this case, the 37° counts are always more variable than the 20°. In comparing chart 5 to chart 4, the wide difference in the means between the 10,000 and the 100,000 dilution results in chart 5 as compared to chart 4 show clearly this point. Among the evenness of melting samples we find the very widest extremes, although there is a certain parallelism between the two dilutions in each sample. On the averages of the triplicate samples from A to F about two-thirds show reasonable uniformity between the two dilutions; the other third shows a wide variation, especially for averages. The averages of the results on samples S to X, in which five different methods were employed, show smaller differences than are to be found among the evenness of melting results; none the less, they are quite wide considering that they are averages of five tests.

CHART NO, 6.

15. Continuing comments 13 and 14 concerning the lack of uniformity in the variations on samples S to X, we find in chart 6 the details of the results obtained at one dilution, 1 to 10,000, at one temperature, 20°C. We note that the results of the different methods of testing show little parallelism. While it would be conceivable that these different methods would not give uniform results on any one sample, we might expect that the difference in method would show the same for each sample, but when we compare the results on sample T with those of samples U or V, we find wide differences. The results with one method are above the fair mean in one sample, and are below on another. In fact, there appears to be no uniformity whatever. This lack of uniformity, therefore, has less to do with the difference in technique than in the composition of the indivdual portion examined by any one method. The variations in the averages in the

individual results by the five methods in samples S to X are greater than the variations in the individual results of the triplicate analyses of samples A to F. In the latter group, the results are more uniform than have been noted in any of the previous charts, and bring us back to the first comment on chart 1.

CHOCOLATE ICE CREAM.—EXPERIMENT NO. 1 AND NO. 3 COMBINED.

Six samples (A, B, C, D, E, F) taken from the same location about 3 inches from the top and at the middle of the can.

Multiply results (except B. coli) by 1,000.

Sample No.	Dilution	Colony Count 37°—2 days B.E.Agar	Total L.L.Agar	Litmus-lactose-agar Acid	Litmus-lactose-agar Inert	Litmus-lactose-agar Alkal.	% Acid Colony Count	Colony Count 20° 5 da. B.E.Agar	Dilution	Gas Production Lac. Pep. Bile 24	48	72	Lit. Lac. Agar Plate	Lac. Agar Stab.	Indol	Nitrate	Gelatine
A1	10,000							8,200	100	—	Tr.	5	+	+	+	+	—
	100,000	27,000	31,400	7,400	24,000		22.2	11,800	1,000	—	—	—					
	1,000,000	30,000						13,000	10,000	—	—	—					
A2	10,000							7,560	100	—	—	5					
	100,000	19,200	19,000	5,200	13,800		27.3	6,800	1,000	—	—	—					
	1,000,000	24,000						9,000	10,000	—	—	—					
A3	10,000							6,300	100	—	—	5					
	100,000	14,600	22,900	6,600	16,300		28.8	7,600	1,000	—	—	Tr.					
	1,000,000	20,000						11,000	10,000	—	—	—					
B1	10,000							6,300	100	—	10	10	+	—	—	+	—
	100,000	23,200	27,000	6,200	20,800		34.0	7,900	1,000	—	Tr.	Tr.	+	—	—	+	—
	1,000,000	23,000						7,000	10,000	—	—	—					
B2	10,000							3,000 spr.	100	—	60	70	+	+	+	+	—
	100,000	27,600	23,700	5,200	18,500		21.9	9,900	1,000	—	—	—					
	1,000,000	40,000						12,000	10,000	—	—	—					
B3	10,000							8,190	100	—	—	—					
	100,000	19,200	31,600	5,600	26,000		17.6	8,900	1,000	—	—	—					
	1,000,000	24,000						8,000	10,000	—	—	—					
C1	10,000							8,820	100	—	Tr.	5	+	+	+	+	—
	100,000	22,900	25,000	6,200	18,800			9,300	1,000	—	—	—					
	1,000,000	18,000						5,000	10,000	—	—	—					
C2	10,000							5,040	100	—	—	—					
	100,000	22,400	33,300	10,000	22,900	400	30.0	8,500	1,000	—	—	—					
	1,000,000	18,000						spreader	10,000	—	—	—					

C3	10,000						8,820	100	—	5	Tr.	+	+	+	+	—
	100,000	26,000	30,200	7,800	22,400	25.8	8,100	1,000	—	—	10					
	1,000,000						5,000	10,000	—	—	—					
D1	10,000						7,560	100	—	30	50	+	+	+	+	—
							1 mould									
	100,000	22,600	34,000	7,200	26,800	24.7	8,000	1,000	—	—	—					
	1,000,000	22,000					10,000	10,000	—	—	—					
D2	10,000						6,930	100	—	—	Tr.					
	100,000	23,200	29,500	7,900	21,600	26.7	8,200	1,000	—	—	Tr.					
	1,000,000	22,000					9,000	10,000	—	—	—					
D3	10,000						8,190	100	—	—	Tr.					
	100,000	18,000	27,400	6,000	21,400	21.9	8,800	1,000	—	—	—					
	1,000,000	13,000					5,000	10,000	—	—	—					
E1	10,000						7,560	100	—	—	Tr.					
	100,000	23,200	30,200	7,000	23,200	23.1	10,100	1,000	—	—	Tr.					
	1,000,000	31,000					7,000	10,000	—	—	—					
E2	10,000						11,340	100	—	Tr.	5	+	+	+	+	—
	100,000	23,600	27,900	7,000	20,900	28.3	7,800	1,000	—	Tr.	10	+	—	—	+	—
	1,000,000	24,000					6,000	10,000	—	—	—					
E3	10,000						6,930	100	—	Tr.	10	+	+	—	—	—
	100,000	19,200	20,900	4,700	16,200	33.5	5,100	1,000	—	—	5					
	1,000,000	31,000					13,000	10,000	—	—	—					
F1	10,000						6,930	100	—	—	Tr.					
	100,000	23,200	24,800	6,200	18,600	25.0	6,800	1,000	—	—	—					
	1,000,000	26,000					15,000	10,000	—	—	—					
F2	10,000						10,710	100	—	Tr.	5					
	100,000	28,000	26,800	8,400	18,400	31.3	10,300	1,000	—	Tr.	10					
	1,000,000	30,000					17,000	10,000	—	—	—					
F3	10,000						6,930	100	—	—	10					
	100,000	27,600	26,800	7,000	19,800	26.1	7,300	1,000	—	—	—					
	1,000,000	34,000					8,000	10,000	—	—	—					

Chocolate Ice Cream Experiments.
Technique of Examinations Same
as Outlined for Experiments on
Vanilla Ice Cream

CHOCOLATE ICE CREAM.—EXPERIMENT NO. 2.

Multiply results (except B. coli) by 1,000.

Description of Sample	Sample No.	Dilution	Colony Count 37°—2 days: B.E.Agar	Colony Count 37°—2 days: Total L.L.Agar	Lactose-Litmus-Agar: Acid	Lactose-Litmus-Agar: Inert	Lactose-Litmus-Agar: Alkal.	% Acid Colony Count	Colony Count 20°-5 da. B.E.Agar	Dilution	Gas Production Lac. Pep. Bile: 24	Gas Production Lac. Pep. Bile: 48	Gas Production Lac. Pep. Bile: 72	Lit. Lac. Agar Plate	Lac. Agar Stab.	Indol	Nitrate	Gelatine
Top Side	G	10,000							8,190	100	—	Tr.	5	+	—	+	+	—
		100,000	31,400	33,100	8,900	24,200		26.8	10,100	1,000	—	—	—					
		1,000,000	32,000						6,000	10,000	—	—	—					
Top Middle	H	10,000							6,930	100	—	10	10	+	+	+	+	—
		1000,000	24,900	28,100	5,900	22,200		21.0	7,900	1,000	—	—	—					
		1,000,000	26,000						12,000	10,000	—	—	—					
Centre Side	I	10,000							10,710	100	Tr.	10	10	+	+	—	+	—
		100,000	38,700	35,800	11,700	23,900	200	32.7	9,200	1,000	—	—	Tr.					
		1,000,000	33,000						12,000	10,000	—	—	—					
Centre Middle	J	10,000							9,450	100	—	—	—					
		100,000	32,700	27,900	7,000	20,600	300	25.0	6,700	1,000	—	—	—					
		1,000,000	18,000						10,000	10,000	—	10	15	‡	‡	=	+	=
Bottom Side	K	10,000							10,080	100	5	60	70	+	+	+	+	—
		100,000	27,200	34,700	7,900	26,800		22.7	10,200	1,000	—	—	Tr.					
		1,000,000	32,000						11,000	10,000	—	—	—					
Bottom Middle	L	10,000							6,300	100	—	—	Tr.					
		100,000	29,800	29,900	7,800	22,000	100	26.0	9,000	1,000	—	—	—					
		1,000,000	28,000						11,000	10,000	—	—	—					

CHOCOLATE ICE CREAM.—EXPERIMENT NO. 5.

Six samples (S, T, U, V, W, X) taken from the same locality at a point adjacent to the place where the six samples for Exp. No. 1 were taken, 3 inches from top near middle of can.

Multiply results (except B. coli) by 1,000.

Sample		Colony Count 37°—2 days									Gas Production							
			Total	Lactose-Litmus-Agar			% Acid	Colony Count			Lac.	Pep.	Bile	Lit. Lac.	Lac.			
No.	Dilution	B.E.Agar	L.L.Agar	Acid	Inert	Alkal.	Colony Count	20°-5 da. B.E.Agar	Dilution		24	48	72	Agar Plate	Agar Stab.	Indol	Nitrate	Gelatine
S1	10,000							6,300	100		+	Tr.	10	+	+	—	+	—
	100,000	24,000	28,700	6,100	22,600		21,2	7,800	1,000		—	—	—					
	1,000,000	28,000						3,000	10,000		—	—	—					
S2	10,000							13,230	100		—	Tr.	5	+	—	—	+	—
	100,000	33,600	37,800	9,500	26,300		25.1	9,900	1,000		—	—	—					
	1,000,000	54,000						6,000	10,000		—	—	—					
S3	10,000							8,820	100		—	10	10	+	+	+	+	—
	100,000	28,200	31,500	7,900	23,600		25.0	8,200	1,000		—	Tr.	5	+	—	—	+	—
	1,000,000	36,000						8,000	10,000		—	—	—					
S4	10,000							10,080	100		—	—	10					
	100,000	37,600	31,300	8,800	22,400	100	28.1	9,100	1,000		—	—	Tr.					
	1,000,000	18,000						14,000	10,000		—	—	Tr.					
S5	10,000							13,800	100		—	60	60	+	+	—	+	—
	100,000	29,900	22,700	5,200	17,500		22.9	7,700	1,000		—	10	10	+	—	—	+	—
	1,000,000	29,000						15,000	10,000		—	—	—					
T1	10,000							7,500	100		—	10	20	—				
	100,000	21,000	19,300	5,200	14,100		26.4	6,500	1,000		—	—	Tr.					
	1,000,000	17,000						6,000	10,000		—	—	—					
T2	10,000							10,710	100		—	—	5					
	100,000	33,500	31,900	10,000	21,900		31.3	8,700	1,000		—	—	Tr.					
	1,000,000	38,000						13,000	10,000		—	—	Tr.					
T3	10,000							8,190	100		—	—	Tr.					
	100,000	17,800	30,300	6,200	24,100		20.4	8,700	1,000		—	—	—					
	1,000,000	30,000						7,000	10,000		—	—	—					
T4	10,000							6,930	100		—	—	Tr.					
	100,000	24,700	22,300	7,500	14,800		21.3	8,200	1,000		—	—	—					
	1,000,000	36,000						10,000	10,000		—	—	—					
T5	10,000							5,670	100		—	—	Tr.					
	100,000	24,900	32,100	11,300	20,800		35.2	6,800	1,000		—	—	—					
	1,000,000	25,000						8,000	10,000		—	—	—					
U1	10,000							10,080	100		—	—	Tr.					
	100,000	25,200	28,200	3,600	24,600		12.0	8,000	1,000		—	—	Tr.					
	1,000,000	24,000						9,000	10,000		—	—	—					
U2	10,000							9,450	100		—	50	50	+	+	+	+	—
	100,000	32,000	45,000	11,700	33,300	200	26.0	9,200	1,000		—	—	—					
	1,000,000	37,000						5,000	10,000		—	—	Tr.					
U3	10,000							7,500	100		Tr.	Tr.	Tr.	+	—	=	+	=
														+	+	—	+	
	100,000	22,300	23,200	7,000	16,200		30.1	7,800	1,000		—	—	Tr.					
	1,000,000	26,000						7,000	10,000		—	—	—					

U4	10,000							9,450	100	—	70	70	+	+	—	+	—
	100,000	24,500	28,700	8,400	20,300		29.2	spread.	1,000	—	—	—					
	1,000,000	30,000						?	10,000	—	—	—					
U5	10,000							10,710	100	—	10	10	+	+	+	+	—
	100,000	20,100		12,700	unc.			6,700	1,000	—	—	—					
	1,000,000	26,000						5,000	10,000	—	—	—					
V1	10,000							7,560	100	—	—	Tr.					
	100,000	30,600	32,200	6,600	25,600		20.5	7,800	1,000	—	—	—					
	1,000,000	46,000						10,000	10,000	—	—	—					
V2	10,000							10,080	100	—	—	Tr.					
	100,000	34,000	43,300	10,300	33,000		23.7	10,200	1,000	—	—	—					
	1,000,000	33,000						11,000	10,000	—	—	Tr.					
V3	10,000							7,560	100	Tr.	30	50	+	—	—	+	—
	100,000	33,400	33,200	10,000	23,200	300	30.1	10,000	1,000	—	—	—					
	1,000,000	32,000						11,000	10,000	—	—	—					
V4	10,000							8,190	100	—	Tr.	10	+	—	+	+	—
	100,000	30,000	28,800	6,800	22,000		23.6	8,800	1,000	—	—	—					
	1,000,000	38,000						10,000	10,000	—	—	—					
V5	10,000							9,450	100	—	10	15	+	+	—	+	—
	100,000	23,000	28,000	6,600	21,400		23.5	8,400	1,000	—	—	Tr.					
	1,000,000	33,000						14,000	10,000	—	—	—					
W1	10,000							10,080	100	—	—	—					
	100,000	26,200	30,700	7,800	22,900		25.4	7,800	1,000	—	—	—					
	1,000,000	30,000						6,000	10,000	—	—	—					
W2	10,000							10,080	100	—	—	Tr.					
	100,000	29,400	31,200	6,600	24,600		20.1	8,200	1,000	—	Tr.	10	—				
	1,000,000	22,000						10,000	10,000	—	—	—					
W3	10,000							11,340	100	—	60	60					
	100,000	35,100	28,800	6,800	22,000		23.7	8,000	1,000	—	—	—					
	1,000,000	31,000						7,000	10,000	—	—	—					
W4	10,000							6,930	100	—	—	Tr.					
	100,000	27,400	33,300	10,300	23,000		30.9	8,100	1,000	—	—	—					
	1,000,000	32,000						11,000	10,000	—	—	—					
W5	10,000							6,300	100	—	—	—					
	100,000	20,500	26,500	6,400	20,100		24.9	5,100	1,000	—	—	—					
	1,000,000	39,000						6,000	10,000	—	—	—					
X1	10,000							9,540	100	—	Tr.	5	+	—	—	+	—
	100,000	29,600	56,600	25,900	30,700		45.7	8,200	1,000	broken							
	1,000,000	28,000						9,000	10,000	—	—	—					
X2	10,000							8,820	100	—	—	Tr.	+	—	—	+	—
	100,000	35,000	65,100	25,300	39,400	400	38.8	9,200	1,000	—	—	Tr					
	1,000,000	33,000						150,000	10,000	—	10	10					
X3	10,000							5,670	100	—	—	—					
	100,000	31,200	67,800	31,200	36,400	200	30.6	9,300	1,000	—	—	5					
	1,000,000	28,000						7,000	10,000	—	—	—					
X4	10,000							9,450	100	—	—	5					
	100,000	28,200		alkaline spreader				7,500	1,000	—	—	—					
	1,000,000	27,000						8,000	10,000	—	—	—					
X5	10,000							10,710	100	—	50	60	+	+	+	+	—
	100,000	26,200	26,100	7,000	19,100		26.8	8,400	1,000	—	—	—					
	1,000,000	23,000						11,000	10,000	—	—	—					

CHOCOLATE ICE CREAM.—EXPERIMENT NO. 4.

Six samples of about 70cc taken from the side of the can from top to bottom.

Multiply results (except B. coli) by 1,000.

Sample No.	Dilution	Colony Count 37°—2 days B.E.Agar	Total L.L.Agar	Litmus-lactose-agar Acid	Inert	Alkal.	% Acid Colony Count	Colony Count 20°-5 da. B.E.Agar	Dilution	Gas Production Lac. Pep. Bile 24	48	72	Lit. Lac. Agar Plate	Lac. Agar Stab.	Indol	Nitrate	Gelatine
M	10,000							7,560	100	—	Tr.	Tr.	+	—	—	+	=
	100,000	2,700	25,600	6.000	19,600		23.4	7,600	1,000	—	—	Tr.					
	1,000,000	3.200						10,000	10,000	—	—	—					
N	10,000							6,930	100	—	5	10	+	—	+	+	—
	100,000	2,600	27,200	6,200	21,000		22.8	8,200	1,000	—	10	10	+	—	—	+	—
	1,000,000	2,900						4,000	10,000	—	—	Tr.					
O	10,000							9,450	100	—	10	20	+	—	—	+	—
	100,000	2.180		6,400	unc.			8,100	1,000	—	—	—					
	1,000,000	2.600						7,000	10,000	—	—	—					
P	10,000							8,820	100	—	10	10	+	+	+	+	—
	100,000	2.720	27,600	6,400	21,200		23.1	7,200	1,000	—	—	—					
	1,000,000	3,000						8,000	10,000	—	Tr.	5	+	—	—	—	—
Q	10,000							6,300	100	—	10	10	+	—	—	+	—
	100,000	2,160	27,000	7,000	20,000		25.9	8,600	1,000	—	—	Tr.					
	1,000,000	2,100						7,000	10,000	—	—	Tr.					
R	10,000							7,560	100	—	—	Tr.					
	100,000	2,300	28,400	6,600	21,800		23.2	8,400	1,000	—	—	—					
	1,000,000	2,000						6,000	10,000	—	—	—					

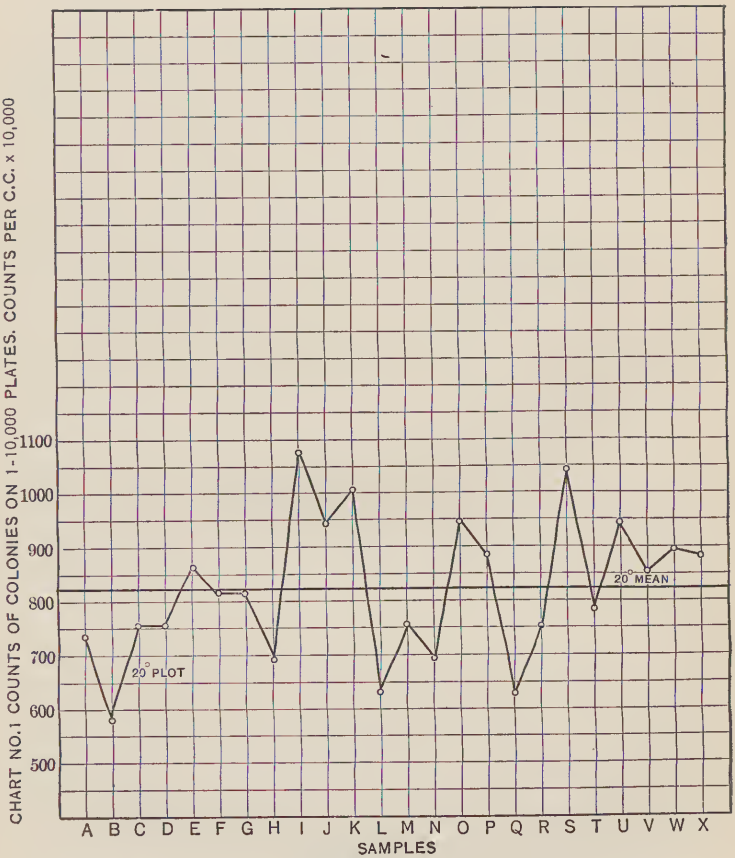


CHART No. 1.

Plot of the 20°C. beef extract agar bacterial counts of Chocolate Ice Cream samples, made from the 1-10,000 dilutions; and of the mean of these counts.

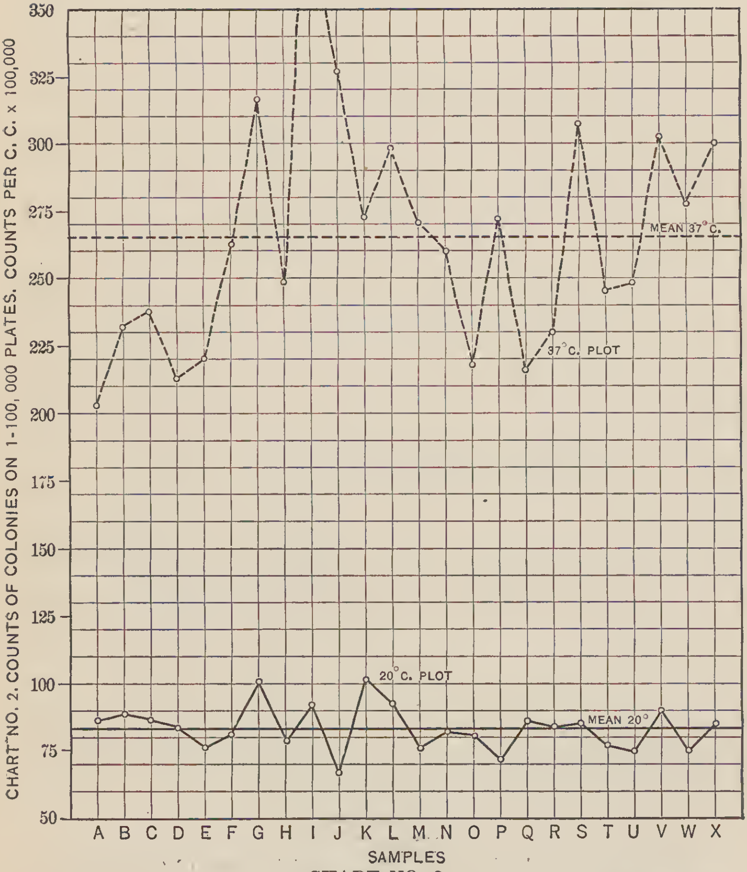


CHART NO. 2.

Plot of the 20°C. and 37°C. beef extract agar bacterial counts of Chocolate Ice Cream samples, made from the 1-100,000 dilutions; and of the means of these counts.

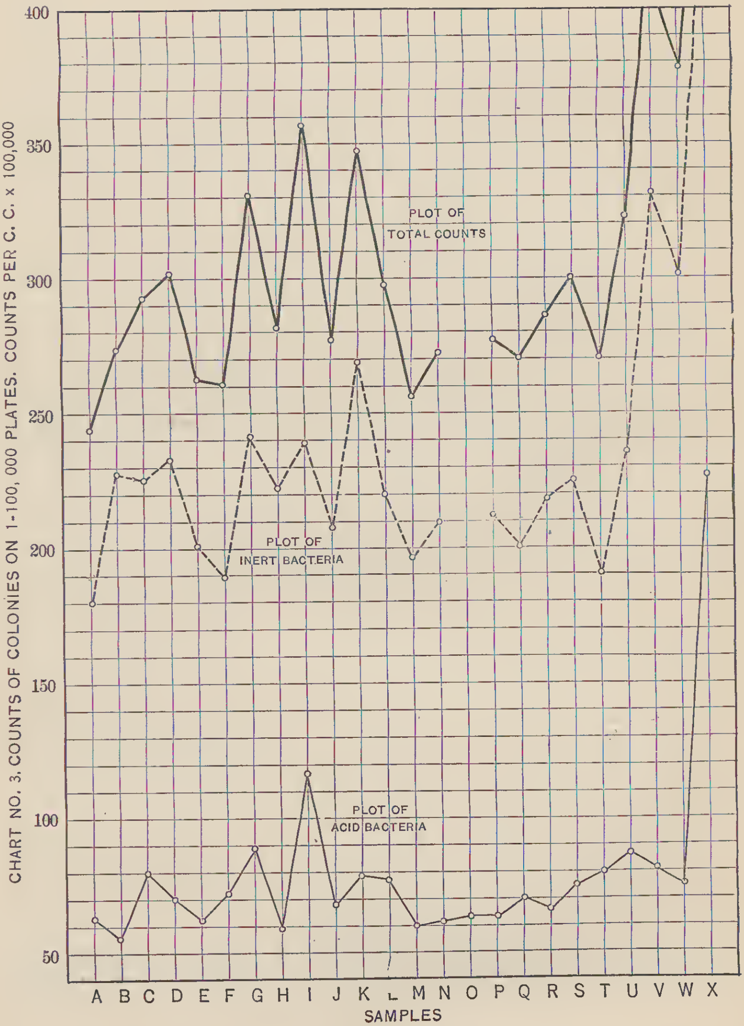


CHART NO. 3.

Plot of the 37°C. litmus lactose agar total and acid forming bacterial counts of Chocolate Ice Cream samples, made from the 1-100,000 dilutions; and of the means of these counts.

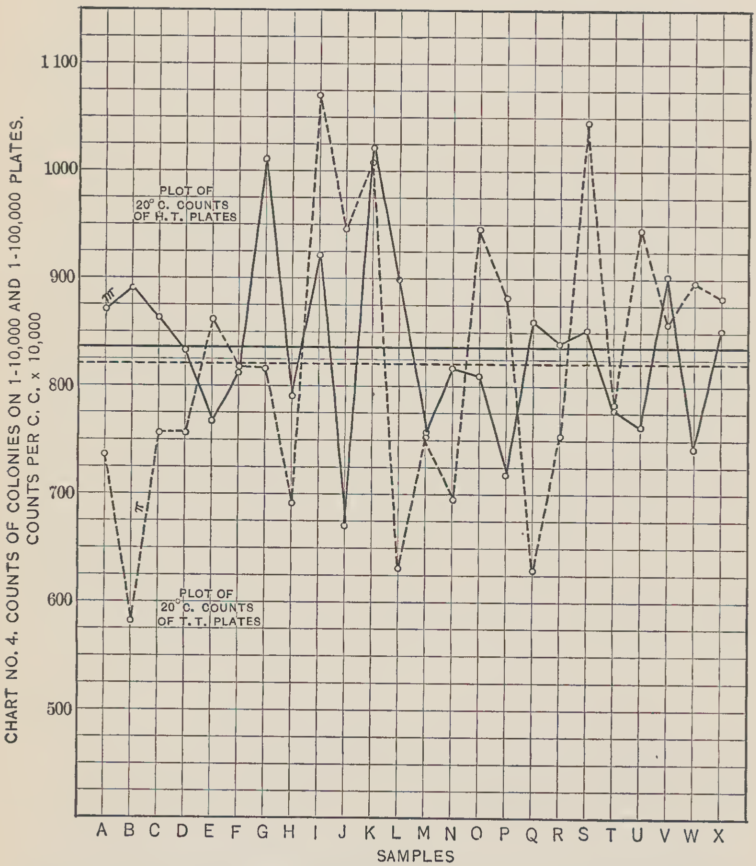


CHART NO. 4.

Plot of the 20°C. beef extract agar bacterial counts of Chocolate Ice Cream samples, made from the 1-10,000 and 1-100,000 dilutions of same samples; and of the means of these counts.

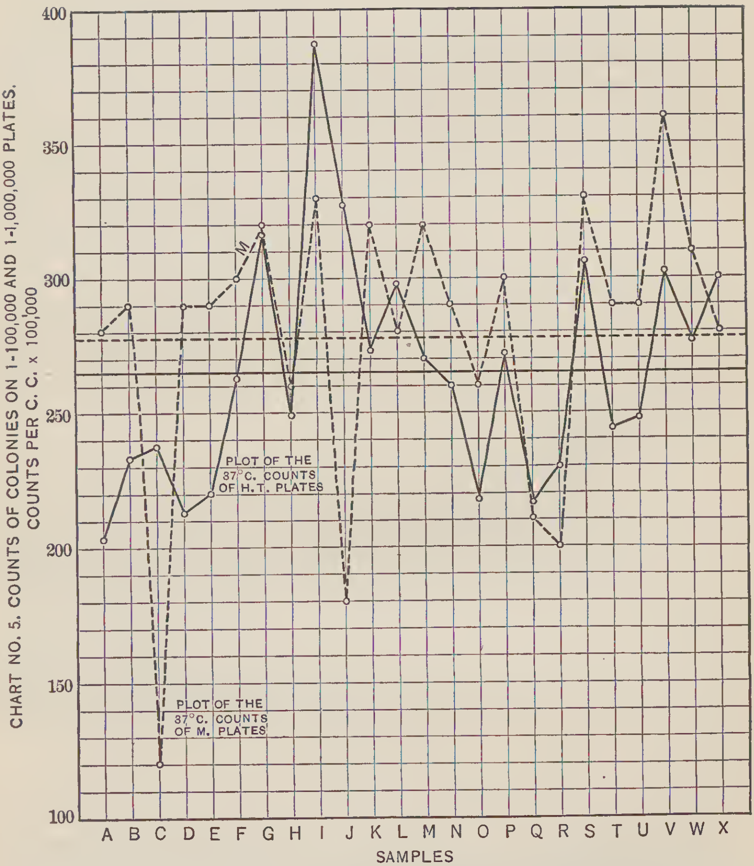


CHART NO. 5.

Plot of the 37°C. beef extract agar bacterial counts of Chocolate Ice Cream samples, made from 1-100,000 and 1-1,000,000 dilutions of the same samples; and of the means of these counts.

Chocolate Ice Cream.

Comments on Results of Tests as Shown by Charts 1 to 5 (Inclusive).

CHART NO. 1.

1. In comparing the results of the averages of triplicate tests on these samples from one location in the can, shown in chart 1, samples A to F, we find differences of somewhat less than 6,000,000 bacteria per c.c. to 8,500,000. For triplicate results, these differences are too great to be explained by anything but an actual difference in the bacterial content of the particular amount of ice cream examined in each test. We find still greater differences when we note the results obtained on individual samples from different portions of the can as shown on the chart on samples G to L (inclusive). Very much the same variations are found among the samples tested for evenness of melting, M to R (inclusive), and we find but little less variation in some of the averages of the five different methods applied to samples S to X.

CHART NO. 2.

2. In this chart we find a picture not previously seen of very wide variation between the means of the 20° and 37° counts. There is a considerable degree of uniformity, however, among the counts for each temperature—a greater degree of uniformity in that respect than we have previously seen. This is especially true of the 20° plottings.

CHART NO. 3.

3. In this chart there is to be noted a wide variation between the total litmus lactose agar counts and the acid forming bacterial counts on the same samples. As in the case of the vanilla ice cream, there is a considerable parallelism between the total counts and the acids counts, as would be expected. This parallelism is even extended to the inert bacteria. In this lot of ice cream the inert bac-

teria played a superior rôle over the acid-forming organisms.

4. A remarkable picture is presented by the variations in the results of different methods of testing samples S to X. Notwithstanding the fact that these samples all came from the same general location, we find an extreme divergence in the averages of the results. This divergence was not noted in the results of the tests at any temperature or any dilution when beef extract agar was used, but appears to be incidental to the use of the litmus lactose agar. It applies in the 1 to 100,000 dilution to both types of organisms present, the inert as well as the acid. This is a good example of the unreliability of litmus lactose agar counts in attempts to determine the sanitary condition of ice cream.

CHART NO. 4.

5. The extreme variations found in the averages of the triplicate tests on samples A to F in this chart are noteworthy. The results on all the other samples in the chart show the usual mixed variability with no sort of uniformity whatever; sometimes the 100,000 plates are higher, and very frequently lower than the 10,000 plates. There is again a series of wide variations in the averages of the S to X sample results.

CHART NO. 5.

6. Again we note the great variability in the averaging of the triplicate samples from the one location in samples from A to F. The 1 to 1,000,000 plates in this chart were on the average higher than the 1 to 100,000 plates, but none the less frequently show lower results than the latter. The extremes of variations in results in this chart, which are the 37° temperature plottings, are greater than the results in the previous chart, which are the 20° tests. Again we find a wide difference in the averages of the

five different methods of testing samples S to X, although there is a certain parallelism between the two dilutions—the 1 to 1,000,000 usually giving the higher results, although in the last sample, X, it fell below.

STRAWBERRY ICE CREAM EXPERIMENTS.

Technique.

Source of Sample. A three-gallon can of Commercial Strawberry Ice Cream was purchased at a retail store and delivered by them to the Lederle Laboratories.

Method of Taking Samples. Sterile teaspoons were used for sampling. The samples were transferred from can to 2-oz. and 4-oz. glass-stoppered bottles.

Three of the samples collected from the same immediate location, 4″ from the surface of the can, in center, were one spoonful only. Three larger samples collected from top, middle and bottom, center of can, were 4 oz. each.

These samples, immediately after removing from can, were packed in ice, and placed in refrigerator until tested.

Method of Melting Sample Before Examination. The smaller samples were melted after taking from ice, by placing in water bath, and shaking thoroughly.

The large 4-oz. samples were allowed to stand at room temperature for ten minutes, and melted portion at bottom of bottle tested, without shaking.

Media. Two batches of all media used were made and duplicate tests conducted, using both sets of media for each sample. One lot of media was made on January 21, 1914, the second lot made on January 28, 1914. The media was stored in refrigerator for ten and three days respectively, before starting experiments.

The following Media were used in examination:

Standard Beef Extract Agar

Standard Inf. Agar.

Lactose Bile (Durham Fermentation tubes).

Standard Casein Agar.

Standard B. E. Agar plus litmus and lactose solutions.

Standard B. E. Agar plus litmus and dextrose solutions.

(All Beef Extract Agar used for making litmus lactose and litmus dextrose plates, was from same lots as that used for making Beef Extract agar plates.)

Dilution Bottles and Diluting Water. The dilution bottles used were 1-oz. and 4-oz. Philadelphia ovals, filled by automatic burettes and to contain 99 c.c. dilution water and 9 c.c. dilution water, respectively after autoclave sterilization. The dilution water contained 0.5% commercial sodium chloride.

Pipettes. The pipettes used were straight sided, single mark and blunt pointed. Capacity pipettes graduated to contain 1 c.c. water, were used for making primary dilution of sample, and volumetric pipettes graduated to deliver 1 c.c. water, for all other dilutions and inoculations.

Petri dishes. Petri dishes 10 x 1.5 cm. sterilized by dry heat at 180°C. for two hours were used for all inoculations (plating).

Method for Making Dilutions and Plating. In all cases, the sample of ice cream was measured by a 1 c.c. single mark capacity pipette, and the pipettes washed out with the dilution water.

The Method of Dilution was as follows:

1 c.c. of sample	to	99	c.c. dilution bottle	= 1:100
1 c.c. of 1:100	to	9	c.c. dilution bottle	= 1:1000
1 c.c. of 1:100	to	99	c.c. dilution bottle	= 1:10000
1 c.c. of 1:10000	to	9	c.c. dilution bottle	= 1:100000
1 c.c. of 1:10000	to	99	c.c. dilution bottle	= 1:1000000

Transfers for higher dilutions were made before 1 c.c. was transferred to petri dish.

Litmus Lactose and Litmus Dextrose Agar Plates. 1 c.c. of a 10% solution of lactose or dextrose, and ½ c.c. of 1% solution of azolitmin were added to each plate before inoculation with 1 c.c. of diluted sample.

Test for Bacteria of B. Coli Type. Durham fermentation tubes were used. The outside tube was 6″ x ⅞″, the inside tube 3″ x ⅜″. The lactose-peptone bile media was made by adding 1% lactose and 1% peptone to fresh ox-bile, which had previously been boiled and filtered.

Two lots of bile media were prepared in accordance with the demands of the experiment and held in the refrigerator until used.

The inoculated tubes were incubated for 72 hours at 37°C., the percentage of gas formed at the end of each 24 hours of incubation being recorded.

From the tubes, litmus lactose agar plates were made at the end of 24, 48 or 72 hours, as gas formation was found.

These plates were made by transferring from tubes, one 3 mm. loopful of bile to a 9 c.c. dilution bottle, and one 3 mm. loopful of this dilution to a plate containing lactose and azolitmin solutions.

After incubation at 37°C. for 24 hours, characteristic colon colonies were isolated from plates to lactose agar slants. These slants were then grown at 37°C. for 24 hours and transfers made from the growths to tubes of Durham's peptone solution, and to nitrate solution.

A stab culture was made in the lactose agar slant, and in a tube of infusion gelatin.

The peptone solution was tested for the presence of indol by the addition of ½ c.c. of paradimethylamidobenyaldehyde solution.

The nitrate solution was tested for the presence of nitrites by the addition of ½ c.c. of a sulphanilic acid solution and ½ c.c. of a naphylamene acetate solution.

The presence of gas in lactose agar stab was noted as a control on the isolation.

The gelatin stabs were incubated 10 days at 20°C., and the presence of liquefaction noted.

Bacterial Counts. All plates were counted, wherever possible.

In reporting actual count per c.c., averages were made in cases where the bacterial count exceeded 20 and was less than 500 colonies to the plate. The number reported was not in accordance with the standard method of reporting bacterial counts.

Outline of Experiments.

To Determine Variations and Bacteriological Data Due to Media Differences.

Experiments were run in duplicate, using the same sample for both tests, but using two lots of media; the first lost being made on January 21st, 1914, and stored in the refrigerator until used January 31st, 1914 (10 days), the second lot being made on January 28th, 1914, and stored in refrigerator until used for test January 31st, 1914 (3 days).

Experiment No. 1. Test of same location.

Three teaspoonful samples: A, B, and C, were collected from the same immediate location, 4″ below surface of cream, and at center of can. These samples were melted in water bath, well shaken, and tested in duplicate for count and coli, using 10-day-old media for one test, and 3-day-old media for other tests.

Experiment No. 2. Test of different locations.

Three samples: (4 oz.) D, E, and F, were collected from top, middle, and bottom of center of can. These samples before testing were allowed to melt 10 minutes at room temperature (70°F.) and the melted portion sampled without shaking, with a 1 c.c. capacity pipette.

STRAWBERRY ICE CREAM.

Multiply results (except B. coli) by 1,000.

BACTERIAL COUNT PER C.C. THREE DAYS AT 37° C. ON

Description of Sample	Sample No.	Dilution	Beef Extract Agar		Dextrose Litmus Agar						Lactose Litmus Agar						Infusion Agar	
			10 days	3 days	10 days Acid	10 days Inert	10 days Alkal.	3 days Acid	3 days Inert	3 days Alkal.	10 days Acid	10 days Inert	10 days Alkal.	3 days Acid	3 days Inert	3 days Alkal.	10 days	3 days
(One teaspoonful each) taken from the same location about 3 inches from the top, in the middle of can.	A	10,000	690	900	580	520	60	810	500	140	490	550	60	410	500	60	350	560
		100,000	400	600	300	100		200	400		500	100		500	500		100	600
		1,000,000	1,000		1,000				1,000		1,000							
	B	10,000	710	790	510	390	90	550	440		420	440	190	370	450	40	280	260
		100,000	600	400	100	300		400	300	100	400	700		100	300	100	500	200
		1,000,000		2,000					1,000		1,000	2,000			3,000		10,000	1,000
	C	10,000	990	970	540	770	70	540	540	70	530	550	50	400	710	20	200	410
		100,000	900	1,000	500	400		500	1,300		500	400		200	500		500	500
		1,000,000		3,000	1,000				1,000		1,000				1,000			1,000
Top Middle	D	10,000	790	590	380	360	90	460	500	40	390	580		420	430	30	170	500
		100,000	500	800	200	600		500	200		200	100		400	900		700	1,100
		1,000,000				spreader		1,000	1,000			1,000			2,000		1,000	2,000
Centre Middle	E	10,000	930	840	480	100	90	370	570	60	430	480		340	500	60	180	440
		100,000	900	700	300	300		1,000	400		400			100	200		300	500
		1,000,000	5,000		1,000			2,000	1,000			1,000			100			
Bottom Middle	F	10,000	830	630	850	80		210	340		280	180		330	500	80	260	380
		100,000	700	400	700	600		100	600		400	100		300	100		300	100
		1,000,000	1,000	3,000	1,000	1,000			2,000								1,000	

Description of Sample	Sample No.	Dilution	THREE DAY BILE									TEN DAY BILE								
			Gas Production Lac. Pep. Bile			Lit. Lac. Agar	Lac. Agar					Gas Production Lac. Lit. Bile			Lit. Lac. Agar	Lac. Agar				
			24	48	72	Plate	Stab.	Nitr.	Pep.	Gela.	B.Coli	24	48	72	Plate	Stab.	Nitrate	Peptone	Gelatine	B.Coli
(One teaspoonful each) taken from the same location about 3 inches from the top, in the middle of can.	A	100	Tr.	40	50	+	+	+	+	—	+	Tr.	60	60	+	+	+	+	—	+
		1,000	—	60	60	+	+	+	+	—	+	—	15	25	+	+	+	+	—	+
		10,000	—	10	10	+	—					—	10	15	+ ?	(=)				
		100,000	—	—	10	+	(=)					—	Tr.	Tr.	—	{ = =				
		1,000,000	—	Tr.	10	+	(=)					—	—	—						
	B	100	Tr.	55	60	+	+	+	+	—	+	10	70	70	+	+	+ sl	+	—	+
		1,000	—	50	60	+	+	+	+	—	+	—	15	15	+	—				
		10,000	—	5	10	+	—					—	10	10	+ ?	(=)				
		100,000	No vials			+ ?	(=)					—	—	—						
		1,000,000	—	—	—							—	—	—						
	C	100	Tr.	50	60	+	+	+	+	—	+	Tr.	50	60	+	+	+	—	—	+
		1,000	—	50	50	+	+	+	+	—	+	—	60	70	+	+	+	+	—	+
		10,000	—	10	15	+	(=)					—	30	70	+	+	+	+	—	+
		100,000	—	Tr.	10	+	(=)					—	10	10	+	(=)				
		1,000,000	—	—	—							—	—	—						
Top Middle	D	100	Tr.	50	60	+	+	+	+	—	+	10	50	50	+	+	+	+	—	+
		1,000	—	70	70	+	+	+	+	—	+	—	80	80	+	+	+	+	—	+
		10,000	—	70	70	+	(‡)	(‡)	(‡)	(=)	(‡)	—	10	10	—	{ (=) +	+ very sl	—	—	+
		100,000	—	—	10	— ?	(=) (=)					—	—	10	—	(=)				
		1,000,000	—	—	—							—	—	—						
Centre Middle	E	100	—	40	50	+	(‡)	(‡)	(‡)	(=)	(‡)	Tr.	50	60	+	+	+	+	—	+
		1,000	—	80	90	+	+	+	+	—	+	—	50	80	+					
		10,000	—	10	20	+ ?	(‡)	(‡)	(=)	(=)	(‡)	—	30	75	+	+	+	+	—	+
		100,000	—	—	—							—	—	10	+	(=)				
		1,000,000	—	—	Tr.	—	(‡)	(‡) sl	(=)	(=)	(‡)	—	—	10	—	(=)				
Bottom Middle	F	100	—	50	60	+	+	+	+	—	+	—	40	50	+	+	+	—	—	+
		1,000	—	90	90	+	+	+	+	—	+	—	50	70	+	+	+	+	—	+
		10,000	—	—	10	— ?	(‡) —	(‡) sl sl	(=)	(‡)	(=)	—	Tr.	10	+ ?	{ = =				
		100,000	—	—	—							—	—	Tr.	+	—				
		1,000,000										—	—	—						

STRAWBERRY ICE CREAM.

Multiply results (except B. coli) by 1,000.

BACTERIAL COUNT PER C.C.

Sample	Dilution	5 days at 20° on Beef Extract Agar 10 da.	3 da.	Infusion Agar 10 da.	3 da.	5 days at 30° on Total Counts 10 da.	3 da.	Casein Agar Peptonizers 10 da.	3 da.
A	10,000	1,430	1,170	900	800	400	440	320	400
	100,000	1,000	1,600	1,100	500	400	500	300	
	1,000,000	4,000	5,000	1,000	1,000	1,000	1,000		
B	10,000	1,360	1,460	600	690	640	480	400	260
	100,000	1,200	700	500	1,100	500	1,000	300	500
	1.000,000	1,000		1.000		2,000		2,000	
C	10,000	1,590	1,210	650	840	560	360	260	220
	100,000	2,000	2,600	1,000	1,000	400	300	300	100
	1.000,000	1,000	1.000		2,000				
D	10,000	1,300	1,050	1.100	1,030	490	300	340	170
	100,000	1,400	1,500	700	400	500	300	500	
	1,000,000	1,000			2,000		1,000		
E	10,000	1.390	1.350	890	800	240	440	190	260
	100,000	1.200	1,100	800	500	400	300	400	200
	1,000,000	5,000	1.000		1,000	1,000	1,000		
F	10,000	1,190	540	560	380	250	370	170	180
	100,000	1.100	500	700	600	200	100	100	
	1,000,000		1.000		1,000	2,000		1,000	

DESCRIPTION OF SAMPLES.

Three samples A, B, C (one teaspoonful each) taken from the same location about 3 inches from the top, in the middle of the can.

D. Top middle. E. Center middle. F. Bottom middle.

Comments on Results of Tests.

The object of these investigations was to determine, as far as one investigation would give results, the effect of different lots of culture media made in the same institution, and by as near as possible the same methods, upon the variability of the bacterial counts. In the investigations of the one lot of strawberry ice cream duplicate culture media were used, one of which had been prepared three days prior to use, and the other ten days. The culture media used included beef extract agar, litmus lactose agar, infusion agar, casein agar, and the usual lactose bile fermentation tube tests for Bacillus coli.

1. There exists no uniform variation between the results from the two lots of culture media. The differences that exist between the counts obtained on any one sample plated on the two media, incubated at any one dilution,

at any one temperature, are no greater, and usually are less, than the differences between different samples taken from immediately contiguous locations in the can when tested on the same culture medium. Differences, therefore, in the preparation of the culture media cannot be a factor which would, in whole or in part, explain the differences of results obtained in the examination of samples from one can of ice cream in any one particular laboratory.

2. It is not even necessary to chart the results of the examinations obtained on this particular lot of ice cream to show the general variability in counts, for the reason that the figures very clearly show very pronounced differences, even when comparisons are made between results of examinations made under identical conditions, as far as possible.

General Conclusions.

1. The results of these investigations indicate conclusively the existence of a great variability in the bacterial counts of different portions of individual cans of ice cream.

2. As a general rule, the bacterial counts obtained by the use of beef extract agar, incubated for five days at 20°C., are subject to less variability than the bacterial counts obtained on the same medium incubated at 37°C. for forty-eight hours.

3. The wide differences noted in the results cannot be accounted for by variations in any of the following details of technique or methods of reading results:

(a) Variations in different lots of beef extract agar, or other media, prepared in the same laboratory under routine conditions.

(b) Slight variations in methods of measurement of the melted ice cream.

(c) By any irregularity in the method of melting the

sample, provided no destruction of bacteria is caused by any method pursued.

(d) By utilizing the results obtained on plates made from dilutions of the melted ice cream yielding either too high or too low counts generally considered satisfactory for general reading.

4. Variations in the bacterial contents of different portions of the can follow no uniform or regular system. One is as likely to find two contiguous samples having the extreme of difference, with widely separated portions showing great uniformity, as the reverse.

5. The making of duplicate plates from any dilution and averaging the results is a waste of effort, for the differences between contiguous samples are far greater than those due to any possible errors in technique.

6. Even the averaging of as many as twenty-four samples from a single can fails to give results which are accurately comparable within narrow limits, although the results are reasonably comparable on a broad basis.

7. From the above conclusions applied broadly to various lots of ice cream, and especially when one considers the detailed relationships between counts at different temperatures, and the different classes of bacteria as to acid and non-acid production and digestion or non-digestion of casein, the conclusion is warranted that each lot of ice cream is a law unto itself.

8. Bacterial counts on ice cream have an important, but very limited, usefulness. They give information only of a general character. It is entirely inappropriate to use them for passing judgment upon a product in any definite manner. The smaller the number of samples taken from a product, the less is the interpreter warranted in utilizing the results as a basis of judgment as to the sanitary character of the product.

9. The conclusions arrived at concerning the value of bacterial counts and their sanitary significance have, to

a large extent, equal applicability to the results of the determination of the numbers of B. coli, as ascertained by the lactose bile fermentation tube method. The standard methods for determining the numbers of B. coli in a given sample tend to develop in the casual observer, or even the technical worker, an unwarranted opinion of the accuracy of the results obtained. The methods do not call for the determination of numbers of B. coli between 100 and 1,000, or 1,000 and 10,000, or 10,000 and 100,-000, etc. Competent bacteriologists have for years understood that any attempt to determine numbers of B. coli with greater accuracy than this method provides would tend to lead to erroneous conclusions. Therefore, reports of numbers of B. coli in samples on any other basis than the numerical system just described tend to lead to fictitious values and should be discountenanced. The results of the examinations of the three lots of ice cream for the numbers of colon bacilli show that even the lactose bile fermentation tube method results are exceedingly variable, and warrant the conclusion that the distribution of these organisms in any one sample of ice cream is far from uniform when they are present in any considerable number.

In the vanilla ice cream, one-half as many samples showed B. coli in 1—1,000 c.c. inoculations as there were in 1—10,000 c.c. inoculations. Approximately one-twelfth of all showed the presence of B. coli in 1—100,000 c.c. inoculations. Thus, a single examination of this ice cream might have given results indicating the presence of either 1,000 or 100,000 B. coli per c.c. Dependence, therefore, upon one test of a sample from a can of ice cream as to the B. coli content would be absolutely valueless. The chocolate ice cream examined contained practically no colon bacilli in 1—100 c.c. inoculations. The strawberry ice cream varied in its B. coli content from 100 B. coli per c.c. to 100,000, although 50% of the tests showed 1,000

B. coli and 33% showed 10,000. These facts show conclusively a totally unscientific basis for attempting to pass judgment upon the sanitary quality of ice cream by attributing sanitary significance to numbers of colon bacilli found in samples of this product, unless one attempted to draw the unwarranted conclusion that an ice cream containing more than a very small number, say 100 colon bacilli per c.c., was not prepared in a sanitary manner.

H. D. PEASE,

Director, Dept. of Bacteriology.

LEDERLE LABORATORIES.

To National Association of Ice Cream Manufacturers.

Bacteriological Investigations Regarding Rates of Growth in Pasteurized Milk

H. D. PEASE, M. D.

REPORTS OF BACTERIOLOGICAL INVESTIGATIONS REGARDING RATES OF GROWTH OF BACTERIA IN PASTEURIZED MILK, PASTEURIZED CREAM, CERTIFIED MILK AND CERTIFIED CREAM REFERRED TO AND PRESENTED IN PART AT THE HEARING.

H. D. Pease, M. D.

Report in the matter of the Examination of Samples of Pasteurized Milk, Pasteurized Cream, Certified Milk and Certified Cream, purchased on the Open Market from Sheffield Farms Slawson-Decker Company, October 28, 1913.

Samples: Six one-quart samples of Certified Milk, six quart samples of Grade "B" Pasteurized Milk were purchased on the open market and received at Lederle Laboratories October 28, 1913.

Method of Obtaining Samples: Without shaking the bottles, 150 c.c. of cream were removed from each of five bottles of certified milk and each of five bottles of pasteurized milk, by means of sterile pipettes. This cream was placed in sterile flasks and packed in ice.

These samples are called certified cream and pasteurized cream respectively.

Method of Examinations: The certified cream and pasteurized cream respectively were placed in 2-oz. sterile bottles, making twelve samples of certified cream and twelve samples of pasteurized cream.

The remaining quarts of certified and pasteurized milk were well shaken, and twelve 2-oz. sterile bottles were filled from each.

These 48 samples were divided into three lots of 16

samples, each lot containing 4 samples of certified cream, certified milk, pasteurized cream, pasteurized milk, one lot being placed at 12°C, one lot at 20°C and one lot at 37°C.

All samples were examined daily, the first examination being of the fresh sample, and the examinations being continued until souring took place.

These examinations were made, for bacterial count, on Beef Extract Agar at 37°C. for 2 days, and for the presence of Colon type organisms in Lactose Peptone Bile at 37°C. for 3 days.

Organoleptic tests were conducted on all samples daily until souring occurred, and finally on the sixth day of holding on all samples.

CERTIFIED CREAM.

Bacteria per c.c. on B.E.Agar, 2 days at 37°C and organoleptic tests. Held at 12°C.

SAMPLE NUMBERS:

Period of Incubation	1		2		3		4	
Fresh	24,000	sweet	31,000	sweet	33,000	sweet	33,000	sweet
1 day	4,800,000	sweet	26,000	sweet	67,000	sweet	27,000	sweet
2 days	23,000	sweet	5,000,000	sweet	32,000,000	sweet	13,000,000	sweet
3 "	15,500	sweet	22,000,000	sweet	6,500,000	sweet	10,000,000	sweet
4 "	unc TT	turning	490,000,000	turning	65,000,000	turning	95,000,000	turning
5 "	no growth M	clabbered	400,000,000	curd whey	28,000,000	turning	300,000,000	turning
6 "		curd whey						

Held at 20°C.

SAMPLE NUMBERS:

Period of Incubation	5		6		7		8	
Fresh	19,000	sweet	1,800	sweet	24,000	sweet	21,000	sweet
1 day	910,000	sweet	710,000	sweet	50,000	sweet	50,000	sweet
2 days	180,000,000	turning	63,000,000	turning	90,000,000	turning	160,000,000	turning
3 "	305,000,000	turning	12,000,000	clabbered	405,000,000	turning	400,000,000	turning
4 "								
5 "								
6 "		curd & whey		curd & whey		curd & whey		curd & whey

Held at 37°C.

SAMPLE NUMBERS:

Period of Incubation	9		10		11		12	
Fresh	27,500	sweet	35,000	sweet	32,000	sweet	40,000	sweet
1 day	560,000,000	turning	140,000,000	turning	560,000,000	turning	440,000,000	turning
2 days	34,500,000	curd whey	30,000,000	curd whey	22,000,000	curd whey	94,000,000	curd whey
3 "								
4 "								
5 "								
6 "								

CERTIFIED MILK.

Bacteria per c.c. on B.E.Agar, 2 days at 37°C and organoleptic tests. Held at 12°C.

SAMPLE NUMBERS:

Period of Incubation	13		14		15		16	
Fresh	1,200	sweet	1,000	sweet	17,000	sweet	3,200	sweet
1 day	900	sweet	900	sweet	20,000	sweet	2,200	sweet
2 days	unc TT spreader TT	sweet	15,000,000	sweet	230,000	sweet	5,000,000	sweet
3 "	24,500,000	sweet	44,000,000	sweet	1,100,000	sweet	2,400,000	sweet
4 "	510,000,000	turning	390,000,000	turning	130,000,000	turning	480,000,000	turning
5 "	1,200,000,000	clabbered	3,000,000,000	clabbered	9,000,000	clabbered	900,000,000	clabbered
6 "		clabbered		curd whey		curd whey		curd whey

Held at 20°C.

SAMPLE NUMBERS:

Period of Incubation	17		18		19		20	
Fresh	900	sweet	420	sweet	16,000	sweet	3,100	sweet
1 day	6,000	sweet	3,000	sweet	40,000	sweet	40,000	sweet
2 days	105,000,000	turning	9,000,000	sweet	29,000,000	sweet	64,000,000	turning
3 "	175,000,000	clabbered	3,900,000	sweet	440,000,000	turning	810,000,000	turning
4 "								
5 "						clabbered		clabbered
6 "		curd whey		clabbered and gas				

Held at 37°C.

SAMPLE NUMBERS:

Period of Incubation	21		22		23		24	
Fresh	1,000	sweet	1,300	sweet	25,000	sweet	2,400	sweet
1 day	755,000,000	sweet	755,000,000	sweet	505,000,000	sweet	440,000,000	sweet
2 days	520,000,000	clabbered	390,000,000	clabbered	215,000,000	clabbered	160,000,000	clabbered
3 "								
4 "								
5 "						curd whey		curd whey
6 "		casein precipitated		casein precipitated				

GRADE B PASTEURIZED CREAM.

Bacteria per c.c. on B.E.Agar, 2 days at 37°C and organoleptic tests. Held at 12°C.

SAMPLE NUMBERS:

Period of Incubation	25		26		27		28	
Fresh	62,000	sweet	74,000	sweet	130,000	sweet	54,000	sweet
1 day	57,000	sweet	20,000 spreader	sweet	20,000	sweet	33,000	sweet (spr.)
2 days	38,000,000	sweet	31,500,000	sweet	24,000,000	sweet	28,000,000	sweet
3 "	310,000,000	sweet	100,000,000	sweet	18,500,000	sweet	170,000,000	sweet
4 "	740,000,000	turning	520,000,000	turning	160,000,000	turning	260,000,000	turning
5 "	50,000,000	curd whey	15,000,000	curd whey	40,000,000	clabbered	180,000,000	clabbered
6 "						curd whey		curd & whey

Held at 20°C.

SAMPLE NUMBERS:

Period of Incubation	29		30		31		32	
Fresh	94,000	sweet	84,000	sweet	90,000	sweet	42,000	sweet
1 day	14,000,000	sweet	12,500,000	sweet	7,800,000	sweet	15,000,000	sweet
2 days	44,000,000	turning	1,100,000,000	turning	650,000,000	turning	1,120,000,000	turning
3 "	40,000,000,000	curd whey	690,000,000	clabbered	265,000,000	turning	230,000,000	turning
4 "				curd whey				
5 "								
6 "						curd whey		curd whey

Held at 37°C.

SAMPLE NUMBERS:

Period of Incubation	33		34		35		36	
Fresh	82,000	sweet	82,000	sweet	120,000	sweet	52,000	sweet
1 day	97,000,000	turning	125,000,000	turning	105,000,000	turning	77,000,000	turning
2 days	74,000,000	curd whey	56,000,000	curd whey	77,000,000	curd whey	12,000,000	curd whey
3 "								
4 "								
5 "								
6 "								

GRADE B PASTEURIZED MILK.

Bacteria per c.c. on B.E.Agar, 2 days at 37°C and organoleptic tests. Held at 12°C.

SAMPLE NUMBERS:

Period of Incubation	37		38		39		40	
Fresh	11,800	sweet	9,800	sweet	60,000	sweet	14,000	sweet
1 day	35,000 spreader	sweet	21,000	sweet	20,000	sweet	21,000	sweet
2 days	11,500,000	sweet	29,000,000	sweet	28,500,000	sweet	40,000,000	sweet
3 "	315,000,000	sweet	175,000,000	sweet	32,000,000	sweet	25,500,000	sweet
4 "	280,000,000	sweet	180,000,000	sweet	235,000,000	sweet	345,000,000	sweet
5 "	80,000,000	turning	14,000,000	turning	20,000,000	turning	70,000,000	turning
6 "		curd whey		clabbered and gas		clabbered		clabbered

Held at 20°C.

SAMPLE NUMBERS:

Period of Incubation	41		42		43		44	
Fresh	31,500	sweet	9,500	sweet	20,000	sweet	No growth TT	sweet
1 day	7,900,000	sweet	10,000,000	sweet	10,800,000	sweet	11,500,000	sweet
2 days	unc H.T. spreader M	turning	820,000,000	turning	940,000,000	turning	1,100,000,000	turning
3 "	380,000,000	clabbered	380,000,000	clabbered	480,000,000	clabbered	450,000,000	clabbered
4 "								
5 "								
6 "		curd & whey		curd & whey		curd & whey		curd & whey

Held at 37°C.

SAMPLE NUMBERS:

Period of Incubation	45		46		47		48	
Fresh	11,000	sweet	8,600	sweet	160,000	sweet	10,000	sweet
1 day	220,000,000	turning	170,000,000	turning	170,000,000	turning	170,000,000	turning
2 days	40,000,000	clabbered	54,000,000	clabbered	62,000,000	clabbered	40,000,000	clabbered
3 "								
4 "								
5 "								
6 "		casein precipitated		casein precipitated		casein precipitated		

CERTIFIED CREAM.

Number of Bacteria of the B.Coli type per c.c. and organoleptic tests. Held at 12°C.

SAMPLE NUMBERS:

Period of Incubation	1		2		3		4	
Fresh	0	sweet	0	sweet	—100	sweet	—100	sweet
1 day	100	sweet	—10	sweet	1,000	sweet	—100	sweet
2 days	1,000 +	sweet	1,000 +	sweet	100,000	sweet	100,000	sweet
3 "	10,000 +	sweet	100,000 +	sweet	100,000	sweet	10,000	sweet
4 "	100,000 +	turning	100,000 +	turning	1,000,000	turning	100,000	turning
5 "	10,000	clabbered	1,000,000	whey curd	100,000	turn. not cl.	1,000,000	turning
6 "		whey curd				turning		turning

Held at 20°C.

SAMPLE NUMBERS:

Period of Incubation	5		6		7		8	
Fresh	0	sweet	0	sweet	—100	sweet	—100	sweet
1 day	10,000	sweet	10,000 +	sweet	10,000	sweet	10,000	sweet
2 days	1,000,000 +	turning	1,000,000 +	turning	1,000,000	turning	1,000,000	turning
3 "	1,000,000 +	turning	1,000,000 +	clabbered	1,000,000 +	turning	1,000,000	turning
4 "								
5 "								
6 "		curd whey		curd whey		curd whey		curd whey

Held at 37°C.

SAMPLE NUMBERS:

Period of Incubation	9		10		11		12	
Fresh	0	sweet	0	sweet	—100	sweet	—100	sweet
1 day	10,000	turning	10,000	turning	100,000	turning	100,000	turning
2 days	—10,000	curd whey	—10,000	curd whey	—1,000,000	curd whey	—10,000	curd whey
3 "								
4 "								
5 "								
6 "								

CERTIFIED MILK.

Number of Bacteria of the B.Coli type per c.c. and organoleptic tests. Held at 12°C.

SAMPLE NUMBERS:

Period of Incubation	13		14		15		16	
Fresh	0	sweet	0	sweet	—100	sweet	—100	sweet
1 day	—10	sweet	—10	sweet	—1,000	sweet	—100	sweet
2 days	1,000	sweet	1,000	sweet	—1,000	sweet	1,000,000	sweet
3 "	1,000	sweet	10,000	sweet	100,000	sweet	100,000	sweet
4 "	100,000	turning	100,000	turning	1,000,000	turning	10,000	turning
5 "	10,000,000	clabbered	1,000,000	clabbered	1,000,000	clabbered	10,000,000	clabbered
6 "		clabbered		curd & whey		curd & whey		curd & whey

Held at 20°C.

SAMPLE NUMBERS:

Period of Incubation	17		18		19		20	
Fresh	0	sweet	0	sweet	—100	sweet	—100	sweet
1 day	—100	sweet	1,000	sweet	—10,000	sweet	—1,000	sweet
2 days	10,000 +	turning	100,000 +	sweet	10,000	sweet	100,000	turning
3 "	10,000 +	clabbered	10,000 +	sweet	1,000,000 +	turning	100,000	turning
4 "								
5 "								
6 "		curd & whey		clabbered + gas		clabbered		clabbered

Held at 37°C.

SAMPLE NUMBERS:

Period of Incubation	21		22		23		24	
Fresh	0	sweet	0	sweet	—100	sweet	—100	sweet
1 day	10,000 +	sweet	10,000 +	sweet	1,000,000	sweet	10,000	sweet
2 days	1,000,000 +	clabbered	10,000	clabbered	—100,000	clabbered	1,000,000	clabbered
3 "								
4 "								
5 "								
6 "		casein precipitated		casein precipitated		curd & whey		curd & whey

GRADE B PASTEURIZED CREAM.

Number of Bacteria of the B.Coli type per c.c. and organoleptic tests. Held at 12°C.

SAMPLE NUMBERS :

Period of Incubation	25		26		27		28	
Fresh	100 +	sweet	100 +	sweet	100	sweet	100	sweet
1 day	100	sweet	1,000 +	sweet	—1,000	sweet	—100	sweet
2 days	10,000 +	sweet	100,000 +	sweet	10,000	sweet	10,000	sweet
3 "	100,000 +	sweet	1,000,000 +	sweet	100,000	sweet	100,000	sweet
4 "	1,000,000 +	turning	100,000	turning	100,000	turning	1,000,000	turning
5 "	—1,000,000	curd & whey	—1,000,000	curd & whey	—1,000,000	clabbered	1,000,000	clabbered
6 "						curd & whey		curd & whey

Held at 20°C.

SAMPLE NUMBERS :

Period of Incubation	29		30		31		32	
Fresh	10	sweet	100	sweet	100	sweet	100	sweet
1 day	10,000 +	sweet	10,000 +	sweet	100,000	sweet	1,000,000	sweet
2 days	10,000,000 +	turning	1,000,000	turning	1,000,000	turning	1,000,000	turning
3 "	10,000,000	curd & whey	1,000,000	clabbered	1,000,000	turning	1,000,000	turning
4 "				curd & whey				
5 "								
6 "						curd & whey		curd & whey

Held at 37°C.

SAMPLE NUMBERS :

Period of Incubation	33		34		35		36	
Fresh	100	sweet	100	sweet	100	sweet	—100	sweet
1 day	100,000	turning	100,000	turning	10,000,000	turning	10,000,000	turning
2 days	10,000,000	curd & whey	1,000,000	curd & whey	—1,000,000	curd & whey	10,000,000	curd & whey
3 "								
4 "								
5 "								
6 "								

GRADE B PASTEURIZED MILK.

Number of Bacteria of the B.Coli type per c.c. and organoleptic tests. Held at 12°C.

SAMPLE NUMBERS:

Period of Incubation	37		38		39		40	
Fresh	10	sweet	10	sweet	—100	sweet	—100	sweet
1 day	10	sweet	100	sweet	—1,000	sweet	100	sweet
2 days	10,000 +	sweet	10,000 +	sweet	100,000	sweet	10,000	sweet
3 "	100,000	sweet	10,000 +	sweet	10,000	sweet	100,000 +	sweet
4 "	1,000,000 +	sweet	100,000	sweet	100,000	sweet	1,000,000	sweet
5 "	1,000,000	turning	100,000	turning	100,000	turning	100,000	turning
6 "		curd & whey		clabbered and gas		clabbered		clabbered

Held at 20°C.

SAMPLE NUMBERS:

Period of Incubation	41		42		43		44	
Fresh	10	sweet	—10.	sweet	—100	sweet	—100	sweet
1 day	10,000	sweet	10,000	sweet	—10,000	sweet	10,000	sweet
2 days	10,000,000 +	turning	1,000,000	turning	100,000	turning	1,000,000	turning
3 "	1,000,000	clabbered	10,000,000 +	clabbered	100,000	clabbered	1,000,000	clabbered
4 "								
5 "								
6 "		curd & whey		curd & whey		curd & whey		curd & whey

Held at 37°C.

SAMPLE NUMBERS:

Period of Incubation	45		46		47		48	
Fresh	1	sweet	10	sweet	—100	sweet	—100	sweet
1 day	100,000	turning	100,000	turning	1,000,000	turning	1,000,000	turning
2 days	1,000,000	clabbered	10,000,000	clabbered	—1,000,000	clabbered	1,000,000	curd & whey
3 "								
4 "								
5 "								
6 "		casein precipitated		casein precipitated		casein precipitated		casein precipitated

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S. L. PARSONS & CO., Inc., Printers, 45 Rose St., New York.